Attorney Docket No.: 81503-AA/GJG/AWG/AC

Prior Application Information

Serial No.: 13/278,333

Class _____ Subclass _____
Examiner: Frank Lu
Group Art Unit: 1634

COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, VA 22313-1450

Date: March 7, 2016

Sir:

This is a request for filing a X continuation _____ divisional _____continuation-in-part application under 37 C.F.R. §1.53(b) of pending application U.S. Serial No. 13/278,333 filed on October 21, 2011 of James Hicks; Nicholas Navin; Jennifer Troge; Zihua Wang; and Michael Wigler for
Inventor(s)

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
Title of Invention
including the following:

X a copy of the prior application (98 pages in total), including 69 pages of specification; 17 page(s) of claims; 1 page(s) of abstract; 0 page(s) of sequence listing; and 11 sheet(s) of drawings)

X Oath or Declaration of Applicant(s) (X signed copy from parent; _____signed new; _____unsigned)

X Power of Attorney (X signed copy from parent; _____signed new; _____unsigned)

X Preliminary Amendment

_____ Amend the specification by inserting before the first line the sentence: --This application is a __continuation __divisional ____continuation-in-part of U.S. Serial No. _____, filed _____, the contents of which are hereby incorporated by reference.--

____ A verified statement to establish small entity status under 37 C.F.R. § 1.27:

____ is enclosed.

____ was filed in the prior application and such status is still proper and desired.

GUARDANT HEALTH EX.1009.001

X The prior application is assigned of record to: Cold Spring Harbor Laboratory, and a copy of the assignment is enclosed.

____ Information Disclosure Statement, including Form PTO-1449 (Copies of citations are included: Yes____; No____)

____ Non-Publication Request, if prior application also had such request (Form PTO/SB/135 must be attached)

____ Computer Readable Form (CRF) of Sequence Listing and Statement Verifying Identity of CRF and Sequence Listing - OR - Request pursuant to 37 C.F.R. §1.821(e) to use the CRF Sequence Listing from the parent.

____ Applicant(s) hereby claim(s) priority based upon the following foreign application(s) under 35 U.S.C. §119

Country	Application No.	Filing Date

____ Certified copy of previously filed foreign application(s) enclosed

____ Certified copy of previously filed foreign application(s) filed in prior application U.S. Serial No. _____, filed on _____.

The filing fee is calculated as follows:

NUMBER OF CLAIMS AS FILED, MINUS ALL CLAIMS CANCELED BY ANY ACCOMPANYING PRELIMINARY AMENDMENT

	Number Filed		Number Extra*				Rate: Small Entity	Rate: Other Entity		Fee: Small Entity	Fee: Other Entity
Total Claims	20	-20 =	0	X			$ 40	$ 80	=	$ 0	$
Independent Claims	3	-3 =	0	X			$ 210	$420	=	$ 0	$
Total Number Of Pages	98 x .75 = 73.50	-100 =	0	÷ by 50	0†	X	$ 200	$400	=	$ 0	$
Multiple Dependent Claims Present: Yes X No							$ 390	$780	=	$ 0	$
Non-English Specification: Yes X No							$ 70	$140	=	$ 0	$
Basic Filing Fee							$ 140	$280	=	$ 70	$
Basic Filing Fee For E-Filing For Small Entity							$ 70		=		
Search Fee									=	$300	$600
Examination Fee									=	$360	$720
Total Fee										$ 730	$

* If less than zero, enter "0"

† Round upwards to integer, e.g.. 1.1 = 2, and insert

____ A check in the amount of $_____ is enclosed.

__X__ Please charge Deposit Account No. 03-3125 in the amount of $ 800.00 .

__X__ The Commissioner is hereby authorized to charge any additional fees required or credit any overpayment to Deposit Account No. 03-3125 as follows:

__X__ Filing fees under 37 C.F.R. §1.16

__X__ Patent application processing fees under 37 C.F.R. §1.17

____ Two copies of this Patent Application Transmittal Letter

____ Return Receipt Postcard

____ Express Mail Certificate of Mailing Label No._____ dated _____

__X__ Other (identify): (1) Application Data Sheet 37 CFR 1.76
(2) Petition under 37 CFR §1.84(a)(2)
(3) $70.00 fee for filing Petition under 37 CFR §1.84(a)(2)

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorneys for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Docket No. 81503-AA/GJG/AWG/AC

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.
U.S. Serial No.:		Not Yet Known (continuation of U.S Serial No. 13/278,333, filed October 21, 2011)
Filed	:	Herewith
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
20th Floor
New York, New York 10112
March 7, 2016

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

Sir:

PRELIMINARY AMENDMENT

This Preliminary Amendment is submitted in connection with the above-identified application prior to the mailing of a first Office Action.

Please amend the subject application as follows:

Amendments to the Claims begin on page **2** of this paper; and

Remarks begin on page **9** of this paper.

Amendments to the Claims

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

72. (New) A method for obtaining genomic copy number information from genomic nucleic acids, comprising:

a) obtaining segments of the genomic nucleic acids;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each tag associated sequence read of step (d); and

f) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining genomic copy number information from the

genomic nucleic acids.

73. (New) The method of claim 72, further comprising estimating a genomic copy number of a region of the genome comprising multiple locations.

74. (New) The method of claim 72, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

75. (New) The method of claim 72, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

76. (New) The method of claim 75, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of

the first region; and

k) obtaining an average of the sum of the counts obtained in step (h) and the counts obtained in step (j).

77. (New) The method of claim 75, wherein the second region of the genome comprises a centromere.

78. (New) The method of claim 72, further comprising l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative genomic copy number of the region of the genome.

79. (New) A method for obtaining mRNA copy number information from mRNA transcripts, comprising:

a) Generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step

(ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information from the mRNA transcripts.

80. (New) The method of claim 72, wherein said tagging the segments comprises:

n) adding a polynucleotide tail to the ends of the segments of the genomic nucleic acids, thereby generating zero-th order

derivative strands comprising the polynucleotide tail;

o) subjecting the zero-th order derivative strands of step (n) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement of the zero-th order derivative strands, thereby generating first order derivative strands;

p) adding a polynucleotide tail to the first order derivative strands, thereby generating a first order derivative strands comprising the polynucleotide tail;

q) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps o) and q) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules.

81. (New) The method of claim 72, wherein said tagging the segments comprises:

r) adding a polynucleotide tail to the ends of the segments of genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail,

s) subjecting the zero-th order derivative strands of step (r) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th

order strands added in step (r) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

t) subjecting the product of step (s) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step r) under conditions which promote the formation of only one complement of the product of step (s), wherein the primers of step (s) and the primers of step (t) have different nucleotide sequences, and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (s) and (t) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, thereby generating tagged nucleic acid molecules.

82. (New) The method of claim 81, wherein said adding the polynucleotide tail comprises the use of a terminal transferase.

83. (New) The method of claim 72, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to step (d).

84. (New) The method of claim 72, wherein the tags further comprise a sample tag.

85. (New) The method of claim 84, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the product of step (c).

86. (New) The method of claim 85, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

87. (New) The method of claim 72, wherein the unique tagged

nucleic acid molecules are generated from a single cell.

88. (New) The method of claim 83, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR in step (c).

89. (New) The method of claim 88, wherein each of the unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array.

90. (New) The method of claim 83, wherein the amplified unique tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to said sequencing the amplified unique tagged nucleic acid molecules in step (d).

91. (New) The method of claim 90, wherein each of the amplified unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on a microarray.

REMARKS

The subject application is a continuation of co-pending U.S Serial No. 13/278,333, filed October 21, 2011. By this Preliminary Amendment, applicants have canceled claims 1-71 without disclaimer or prejudice to applicant's right to pursue the subject matter of these claims in a related application. Applicants have also added new claims 72-91. Upon entry of this Preliminary Amendment, claims 72-91 will be pending.

Support for new claim 72 may be found, *inter alia*, in the publication of the '333 parent application at page 1, paragraphs [0011]-[0018], page 9, paragraphs [0274]-[0275], and page 10, paragraph [0291].

Support for new claim 73 may be found, *inter alia*, in the publication of the '333 parent application at page 4, paragraph [0106].

Support for new claim 74 may be found, *inter alia*, in the publication of the '333 parent application at page 4, paragraph [0107].

Support for new claim 75 may be found, *inter alia*, in the publication of the '333 parent application at page 4, paragraphs [0108-112].

Support for new claim 76 may be found, *inter alia*, in the publication of the '333 parent application at page 4, paragraphs [0113-115].

Support for new claim 77 may be found, *inter alia*, in the

publication of the '333 parent application at page 4, paragraph [0116].

Support for new claim 78 may be found, *inter alia*, in the publication of the '333 parent application at page 4, paragraph [0117].

Support for new claim 79 may be found, *inter alia*, in the publication of the '333 parent application at page 2, paragraphs [0029]-[0038].

Support for new claims 80, 81 and 82 may be found, *inter alia*, in the publication of the '333 parent application at page 3, paragraphs [0090]-[0093], Figures 8 and 9, page 5, paragraphs [0150]-[0155] and pages 11-13 , paragraphs [0303]-[0320].

Support for new claims 83 and 88-91 may be found, *inter alia*, in the publication of the '333 parent application at page 5, paragraph [0172], page 6, paragraph [0218], page 7, paragraph [0246] and page 14, paragraph [0330].

Support for new claims 84-87 may be found, *inter alia*, in the publication of the '333 parent application at page 8, paragraph [0266], page 9, paragraph [0278], page 10, paragraph [0283] and page 11, paragraph [0303].

Applicants maintain that the amendments to the claims raise no issue of new matter. Accordingly, applicants respectfully request entry of this Preliminary Amendment.

If a telephone conference would be of assistance in advancing prosecution of this application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the enclosed filing fee of $730.00 and a $70.00 fee for filing an accompanying Petition under 37 CFR § 1.84(a)(2) to Accept Color Drawings, is deemed necessary in connection with the filing of this Preliminary Amendment and the accompanying application. However, if any additional fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik

Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmittal

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

3/7/16

Andrew W. Gorman Date

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

The application data sheet is part of the provisional or nonprovisional application for which it is being submitted. The following form contains the bibliographic data arranged in a format specified by the United States Patent and Trademark Office as outlined in 37 CFR 1.76.
This document may be completed electronically and submitted to the Office in electronic format using the Electronic Filing System (EFS) or the document may be printed and included in a paper filed application.

Secrecy Order 37 CFR 5.2:

☐ Portions or all of the application associated with this Application Data Sheet may fall under a Secrecy Order pursuant to 37 CFR 5.2 (Paper filers only. Applications that fall under Secrecy Order may not be filed electronically.)

Inventor Information:

Inventor 1 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	James		Hicks	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	Locust Valley	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	3 Great Meadows Road
Address 2	

City	Locust Valley	State/Province	NY
Postal Code	11560	Country	US

Inventor 2 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Nicholas		Navin	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	Houston	State/Province	TX	Country of Residence	US

Mailing Address of Inventor:

Address 1	1621 Wichita Street
Address 2	

City	Houston	State/Province	TX
Postal Code	77004	Country	US

Inventor 3 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Jennifer		Troge	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

City	Oyster Bay	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	100 Simcoe Street		
Address 2			
City	Oyster Bay	State/Province	NY
Postal Code	11771	Country	US

Inventor 4 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Zihua		Wang	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	Huntington	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	13 Meadowlawn Street		
Address 2			
City	Huntington	State/Province	NY
Postal Code	11743	Country	US

Inventor 5 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Michael		Wigler	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	Cold Spring Harbor	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	52 Snake Hill Road		
Address 2			
City	Cold Spring Harbor	State/Province	NY
Postal Code	11724	Country	US

All Inventors Must Be Listed - Additional Inventor Information blocks may be generated within this form by selecting the **Add** button. Add

Correspondence Information:

Enter either Customer Number or complete the Correspondence Information section below.
For further information see 37 CFR 1.33(a).

☐ **An Address is being provided for the correspondence Information of this application.**

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Customer Number	23432		
Email Address	patentdocketing@cooperdunham.com	Add Email	Remove Email

Application Information:

Title of the Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION		
Attorney Docket Number	81503-AA/GJG/AWG/AC	**Small Entity Status Claimed**	☒
Application Type	Nonprovisional		
Subject Matter	Utility		
Total Number of Drawing Sheets (if any)	11	**Suggested Figure for Publication (if any)**	

Filing By Reference:

Only complete this section when filing an application by reference under 35 U.S.C. 111(c) and 37 CFR 1.57(a). Do not complete this section if application papers including a specification and any drawings are being filed. Any domestic benefit or foreign priority information must be provided in the appropriate section(s) below (i.e., "Domestic Benefit/National Stage Information" and "Foreign Priority Information").

For the purposes of a filing date under 37 CFR 1.53(b), the description and any drawings of the present application are replaced by this reference to the previously filed application, subject to conditions and requirements of 37 CFR 1.57(a).

Application number of the previously filed application	Filing date (YYYY-MM-DD)	Intellectual Property Authority or Country

Publication Information:

☐ Request Early Publication (Fee required at time of Request 37 CFR 1.219)

☐ **Request Not to Publish.** I hereby request that the attached application not be published under 35 U.S.C. 122(b) and certify that the invention disclosed in the attached application **has not and will not** be the subject of an application filed in another country, or under a multilateral international agreement, that requires publication at eighteen months after filing.

Representative Information:

Representative information should be provided for all practitioners having a power of attorney in the application. Providing this information in the Application Data Sheet does not constitute a power of attorney in the application (see 37 CFR 1.32). Either enter Customer Number or complete the Representative Name section below. If both sections are completed the customer Number will be used for the Representative Information during processing.

Please Select One:	◉ Customer Number	○ US Patent Practitioner	○ Limited Recognition (37 CFR 11.9)
Customer Number	23432		

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Domestic Benefit/National Stage Information:

This section allows for the applicant to either claim benefit under 35 U.S.C. 119(e), 120, 121, 365(c), or 386(c) or indicate National Stage entry from a PCT application. Providing benefit claim information in the Application Data Sheet constitutes the specific reference required by 35 U.S.C. 119(e) or 120, and 37 CFR 1.78.
When referring to the current application, please leave the "Application Number" field blank.

Prior Application Status	Pending		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
	Continuation of	13/278333	2011-10-21
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/510579	2011-07-22
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/406067	2010-10-22

Additional Domestic Benefit/National Stage Data may be generated within this form by selecting the **Add** button.

Foreign Priority Information:

This section allows for the applicant to claim priority to a foreign application. Providing this information in the application data sheet constitutes the claim for priority as required by 35 U.S.C. 119(b) and 37 CFR 1.55. When priority is claimed to a foreign application that is eligible for retrieval under the priority document exchange program (PDX)[i] the information will be used by the Office to automatically attempt retrieval pursuant to 37 CFR 1.55(i)(1) and (2). Under the PDX program, applicant bears the ultimate responsibility for ensuring that a copy of the foreign application is received by the Office from the participating foreign intellectual property office, or a certified copy of the foreign priority application is filed, within the time period specified in 37 CFR 1.55(g)(1).

			Remove
Application Number	Country[i]	Filing Date (YYYY-MM-DD)	Access Code[i] (if applicable)

Additional Foreign Priority Data may be generated within this form by selecting the **Add** button.

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

☐ This application (1) claims priority to or the benefit of an application filed before March 16, 2013 and (2) also contains, or contained at any time, a claim to a claimed invention that has an effective filing date on or after March 16, 2013.
NOTE: By providing this statement under 37 CFR 1.55 or 1.78, this application, with a filing date on or after March 16, 2013, will be examined under the first inventor to file provisions of the AIA.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Authorization or Opt-Out of Authorization to Permit Access:

When this Application Data Sheet is properly signed and filed with the application, applicant has provided written authority to permit a participating foreign intellectual property (IP) office access to the instant application-as-filed (see paragraph A in subsection 1 below) and the European Patent Office (EPO) access to any search results from the instant application (see paragraph B in subsection 1 below).

Should applicant choose not to provide an authorization identified in subsection 1 below, applicant **must opt-out** of the authorization by checking the corresponding box A or B or both in subsection 2 below.

NOTE: This section of the Application Data Sheet is **ONLY** reviewed and processed with the **INITIAL** filing of an application. After the initial filing of an application, an Application Data Sheet cannot be used to provide or rescind authorization for access by a foreign IP office(s). Instead, Form PTO/SB/39 or PTO/SB/69 must be used as appropriate.

1. Authorization to Permit Access by a Foreign Intellectual Property Office(s)

A. Priority Document Exchange (PDX) - Unless box A in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the European Patent Office (EPO), the Japan Patent Office (JPO), the Korean Intellectual Property Office (KIPO), the State Intellectual Property Office of the People's Republic of China (SIPO), the World Intellectual Property Organization (WIPO), and any other foreign intellectual property office participating with the USPTO in a bilateral or multilateral priority document exchange agreement in which a foreign application claiming priority to the instant patent application is filed, access to: (1) the instant patent application-as-filed and its related bibliographic data, (2) any foreign or domestic application to which priority or benefit is claimed by the instant application and its related bibliographic data, and (3) the date of filing of this Authorization. See 37 CFR 1.14(h)(1).

B. Search Results from U.S. Application to EPO - Unless box B in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the EPO access to the bibliographic data and search results from the instant patent application when a European patent application claiming priority to the instant patent application is filed. See 37 CFR 1.14(h)(2).

The applicant is reminded that the EPO's Rule 141(1) EPC (European Patent Convention) requires applicants to submit a copy of search results from the instant application without delay in a European patent application that claims priority to the instant application.

2. Opt-Out of Authorizations to Permit Access by a Foreign Intellectual Property Office(s)

☐ A. Applicant **DOES NOT** authorize the USPTO to permit a participating foreign IP office access to the instant application-as-filed. If this box is checked, the USPTO will not be providing a participating foreign IP office with any documents and information identified in subsection 1A above.

☐ B. Applicant **DOES NOT** authorize the USPTO to transmit to the EPO any search results from the instant patent application. If this box is checked, the USPTO will not be providing the EPO with search results from the instant application.

NOTE: Once the application has published or is otherwise publicly available, the USPTO may provide access to the application in accordance with 37 CFR 1.14.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Applicant Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

Applicant 1

If the applicant is the inventor (or the remaining joint inventor or inventors under 37 CFR 1.45), this section should not be completed. The information to be provided in this section is the name and address of the legal representative who is the applicant under 37 CFR 1.43; or the name and address of the assignee, person to whom the inventor is under an obligation to assign the invention, or person who otherwise shows sufficient proprietary interest in the matter who is the applicant under 37 CFR 1.46. If the applicant is an applicant under 37 CFR 1.46 (assignee, person to whom the inventor is obligated to assign, or person who otherwise shows sufficient proprietary interest) together with one or more joint inventors, then the joint inventor or inventors who are also the applicant should be identified in this section.

Clear

◯ Assignee	◯ Legal Representative under 35 U.S.C. 117	◯ Joint Inventor
◯ Person to whom the inventor is obligated to assign.	◯ Person who shows sufficient proprietary interest	

If applicant is the legal representative, indicate the authority to file the patent application, the inventor is:

Name of the Deceased or Legally Incapacitated Inventor:

If the Applicant is an Organization check here. ☐

Prefix	**Given Name**	Middle Name	**Family Name**	Suffix

Mailing Address Information For Applicant:

Address 1			
Address 2			
City		**State/Province**	
Country		Postal Code	
Phone Number		Fax Number	
Email Address			

Additional Applicant Data may be generated within this form by selecting the Add button.

Assignee Information including Non-Applicant Assignee Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Assignee 1

Complete this section if assignee information, including non-applicant assignee information, is desired to be included on the patent application publication. An assignee-applicant identified in the "Applicant Information" section will appear on the patent application publication as an applicant. For an assignee-applicant, complete this section only if identification as an assignee is also desired on the patent application publication.

If the Assignee or Non-Applicant Assignee is an Organization check here. ☒

Organization Name	Cold Spring Harbor Laboratory

Mailing Address Information For Assignee including Non-Applicant Assignee:

Address 1	One Bungtown Road		
Address 2			
City	Cold Spring Harbor	**State/Province**	NY
Country i	US	Postal Code	11724
Phone Number		Fax Number	
Email Address			

Additional Assignee or Non-Applicant Assignee Data may be generated within this form by selecting the Add button.

Signature:

NOTE: This Application Data Sheet must be signed in accordance with 37 CFR 1.33(b). **However, if this Application Data Sheet is submitted with the INITIAL filing of the application and either box A or B is not checked in subsection 2 of the "Authorization or Opt-Out of Authorization to Permit Access" section, then this form must also be signed in accordance with 37 CFR 1.14(c).**

This Application Data Sheet **must** be signed by a patent practitioner if one or more of the applicants is a **juristic entity** (e.g., corporation or association). If the applicant is two or more joint inventors, this form must be signed by a patent practitioner, **all** joint inventors who are the applicant, or one or more joint inventor-applicants who have been given power of attorney (e.g., see USPTO Form PTO/AIA/81) on behalf of **all** joint inventor-applicants.

See 37 CFR 1.4(d) for the manner of making signatures and certifications.

Signature	/Gary J. Gershik/			Date (YYYY-MM-DD)	2016-03-07
First Name	Gary	Last Name	Gershik	Registration Number	39992

Additional Signature may be generated within this form by selecting the Add button.

3/7/16

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

This collection of information is required by 37 CFR 1.76. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 23 minutes to complete, including gathering, preparing, and submitting the completed application data sheet form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1 The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.

2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.

3 A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.

4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).

5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent CooperationTreaty.

6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).

7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.

8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.

9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

GUARDANT HEALTH EX.1009.024

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

This application claims the benefit of U.S. Provisional Application Nos. 61/510,579, filed July 22, 2011, and 61/406,067, filed October 22, 2010 the contents of each of which are hereby incorporated by reference in their entirety.

The invention disclosed herein was supported by the government under grant No. W81XWH-09-1-0591 from the Department of Defense. Accordingly, the U. S. Government has certain rights to this invention.

Throughout this application, various publications are referenced by numbers in parentheses. Full citations for these references may be found at the end of the specification immediately preceding the claims. The disclosures of these publications in their entireties are hereby incorporated by reference into this application to more fully describe the state of the art to which this invention pertains.

Background of the Invention

Genomic copy number information is commonly obtained using whole genome amplification (WGA). The endemic problem with the WGA method is over-sampling of certain regions, yielding a non-uniform amplification of the genome (1). WGA methods begin with the step that initiates the process, a polymerase (Phi 29) makes a strand from genomic DNA utilizing a random primer coupled to an adaptor for subsequent PCR (Figure 1). If the input DNA strands are referred to as the "0-th derivative", and the first synthesized strand as a "first derivative," subsequent strands are called the (n+1)-th derivative if their template was an n-th derivative. Only strands that are 2-nd derivative or higher become amplified in the PCR step,

resulting in a 'stacking' over the regions 'chosen' by the polymerase for the first derivative.

Coverage of the genome by sequencing WGA of single cell DNA is limited by the stacking phenomenon (Figure 2). Thus it is difficult to obtain single cell measurements, particularly when based on WGA, due to distortions that originate from stochastic sampling and amplification steps. Moreover, the current method of WGA is a black box, with the unspecified reagents purchased from a vendor, which hampers optimization. Moreover, the WGA method does not extend to a method usable for single cell RNA profiling.

Ligation-mediated PCR was developed in an attempt to solve the above-identified problems inherent in WGA. In this method, adaptors are ligated to an *Mse*I restriction endonuclease digest of genomic DNA from a single cell, followed by PCR amplification using primers complementary to the adaptors. The amplified DNA is then used for CGH or DNA sequencing (2,3). However, like WGA, the method still requires an amplification step.

Parameswaran et al. (2007) and U.S. Patent No. 7,622,281 describe methods of labeling nucleic acid molecules with barcodes for the purpose of identifying the source of the nucleic acid molecules, thereby allowing for high-throughput sequencing of multiple samples (4,5). Eid et al. (2009) describe a single molecule sequencing method wherein single-molecule real time sequencing data is obtained from a DNA polymerase performing uninterrupted template-directed synthesis using four distinguishable fluorescently labeled dNTPs (6). However, these methods do not provide genomic information unaffected by amplification distortion.

Miner et al. (2004) describe a method of molecular barcoding to label template DNA prior to PCR amplification, and report that

the method allows for the identification of contaminant and redundant sequences by counting only distinctly tagged sequences (22). U.S. Patent No. 7,537,897 describes methods for molecular counting by labeling molecules of an input sample with unique oligonucleotide tags and subsequently amplifying and counting the number of different tags (23). Miner et al. and U.S. Patent No. 7,537,897 both describe labeling of input nucleic acid molecules by ligation, which has been found to be an inefficient reaction.

McCloskey et al. (2007) describe a method of molecular encoding which does not use ligation but instead uses template specific primers to barcode template DNA molecules prior to PCR amplification (24). However, such a method requires that template specific primers be made for each species of template DNA molecule studied.

As described herein, obtaining accurate genomic copy number information by high-throughput sequencing of genomic DNA prepared by WGA methods is hampered by the copy number distortions introduced by non-uniform amplification of genomic DNA. Thus, there exists a need for a method that allows for copy number determination free of distortions caused by amplification steps and which allows for accurate and efficient copy number determination of complex samples. Such a method should also be robust using existing methodologies for high volume, massively parallel sequencing.

SUMMARY OF THE INVENTION

A method is provided for obtaining from genomic material genomic copy number information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) tagging the segments with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises one segment of the genomic material from step (a) and a tag;

c) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

d) generating tag associated sequence reads by sequencing the product of step (c);

e) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome; and

f) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining genomic copy number information unaffected by amplification distortion.

Also provided is a method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA;

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises

i) one cDNA from step (a) or one segment of the cDNA from step (b), and

ii) a tag;

d) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

e) generating tag associated sequence reads by sequencing the product of step (d);

f) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a cDNA or a segment of cDNA to a location on the cDNA library; and

g) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

Also provided is a method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) generating tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to the first order derivative strands; and

iii) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise substantially unique primers, thereby generating tagged nucleic acid molecules;

b) subjecting the tagged nucleic acid molecules to polymerase chain reaction (PCR) amplification;

c) generating tag associated sequence reads by sequencing the product of step (b);

d) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a mRNA transcript to a location on the cDNA library; and

e) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

Also provided is a method for obtaining from genomic material DNA methylation information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands to a polymerase reaction in the presence essentially unique primers, wherein the essentially unique primers are capable of hybridizing to the polynucleotide tail of the zero-th order strands, under conditions that promote the formation of only one complement, thereby generating tagged nucleic acid molecules;

d) separating the tagged nucleic acid molecules into a group consisting of hemi-methylated

tagged nucleic acid molecules and a group consisting of unmethylated tagged nucleic acid molecules;

e) subjecting each group of step (d) to amplification by polymerase chain reaction (PCR);

f) generating tag associated sequence reads by sequencing the product of step (e);

g) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome; and

h) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining DNA methylation information unaffected by amplification distortion.

Also provided is a composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands of step (b) to a polymerase reaction in the presence of primers capable of

hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

d) adding a polynucleotide tail to the first order derivative strands;

e) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (c) and (e) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

Also provided is a composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of genomic material;

b) adding a polynucleotide tail to the ends of the segments of genomic material to generate zero-th order derivative strands,

c) subjecting the zero-th order derivative strands of step (b) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (b) under

conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

d) subjecting the product of step (c) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (b) under conditions which promote the formation of only one complement, wherein the primers of step (d) have different nucleotide sequences than the primers of step (c), and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (c) and (d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

Also provided is a composition of matter derived from a mRNA transcripts comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA; and

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, wherein each tagged nucleic acid molecule comprises

i) one cDNA from step (a) or one segment of the cDNA from step (b) and

ii) a tag.

Also provided is a composition of matter derived from mRNA transcripts comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining mRNA transcripts;

b) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

c) adding a polynucleotide tail to the first order derivative strands;

d) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (c) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (b) and d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

Also provided is a kit for determining nucleic acid copy number information unaffected by amplification distortion comprising:

a) a terminal transferase, and

b) plurality of substantially unique primers,

wherein the substantially unique primers comprise substantially unique tags, and wherein the substantially unique primers are capable of hybridizing to the polynucleotide tail of a nucleic acid molecule which has a polynucleotide tail added by the terminal transferase.

BRIEF DESCRIPTION OF THE FIGURES

The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawing(s) will be provided by the Office upon request and payment of the necessary fee.

Figure 1. Whole Genome Amplification (WGA).

Figure 2. Genome coverage comparison.

Genomic coverage comparison of 7 sequencing lanes from one single cell library versus 7 lanes from a million cell library. Stacking causes the single cell reads to be concentrated over the first derivative reads from WGA. More reads yield diminishing returns.

Figure 3. Sequencing read counts for CpG dinucleotides within selected CpG islands

Normal cell line SKN-1 (A,C,E) and breast cancer line MDA-MB-231 (B,D,F). Orange indicates T residues (unmethylated). Blue indicates C residues (methylated). Counts of the plus strand are above the X-axis, and counts on the minus strand are below.

Figure 4. Comparison of array CGH with 'sequence counting' for analyzing copy number data.

Graph shows a portion of chromosome 17 from FFPE sample JZ33 including raw (gray) ratio and segmented (green) data from array CGH, and raw sequence count data (orange) and its segmentation (blue).

Figure 5. ROMA CGH of sKBR3 compared with profiles of 3 independent single cell isolates derived from sparse DNA sequencing.

Figure 6. Sequencing across the breakpoints of a homozygous deletion in cell on chromosome 5 line T47D.

Figure 7. Flowchart for genomic profiling by representational sequence counting

Figure 8. Scheme for varietal counting. Varietal tags are shown as random nucleotide sequences (N_{10})

Flowchart of one embodiment of the invention.

Figure 9. Nick ligation protocol

The sequence of primer A includes universal primer A (PrA; green), varietal tag (B; yellow), and oligo dT+CATG, the sequence of primer B includes universal primer B (PrB; orange) and oligo dT+CATG, PrA and PrB are universal primers, and cPrA(blue), cPrB (magenta), and cB(blue) are complementary to PrA, PrB and B, respectively.

Figure 10. Schema for targeted sequencing of regional markers

UfBPrA is forward primer, B (yellow) is tag, UrPrR is reverse primer, PrB (brown) is gene-specific reverse primer, Uf (orange) is universal forward primer, PrA (blue) is gene specific forward primer, Ur (magenta) is universal reverse primer.

Figure 11. Comparison of Varietal Counting with Sequence Counting

Figure 11A shows genome wide copy number data. Figure 11B shows partial genome copy number data.

DETAILED DESCRIPTION OF THE INVENTION

A method is provided for obtaining from genomic material genomic copy number information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) tagging the segments with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises one segment of the genomic material from step (a) and a tag;

c) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

d) generating tag associated sequence reads by sequencing the product of step (c);

e) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome; and

f) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining genomic copy number information unaffected by amplification distortion.

In an embodiment, the method further comprises estimating a genomic copy number of a region of the genome comprising more than one location on the genome by assigning as the copy

number of the region the highest count obtained in step (f) for the locations within the region.

In an embodiment, the method further comprises comparing a count obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

In an embodiment, the method further comprises

a) summing the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

b) summing the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region is comprised of a number of locations which is comparable to the number of locations of the first region;

c) comparing the value obtained in step (a) to the value obtained in step (b),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

Step (b) of the above embodiment may further comprise

i) summing the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region is comprised of a number of locations which is comparable to the

number of locations of the first region; and

ii) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (f) for locations which comprise the third region.

In an embodiment of the method, the second region of the genome comprises a centromere.

In an embodiment, the method further comprises summing the counts obtained in step (f) for locations which comprise a region of the genome, and comparing the sum to a sum obtained from a reference sample for the same region of the genome, thereby estimating a relative genomic copy number of the region of the genome.

Also provided is a method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA;

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises

i) one cDNA from step (a) or one segment of the cDNA from step (b), and

ii) a tag;

d) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

e) generating tag associated sequence reads by sequencing the product of step (d);

f) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a cDNA or a segment of cDNA to a location on the cDNA library; and

g) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

Also provided is a method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) generating tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to the first order derivative strands; and

iii) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise substantially unique primers, thereby generating tagged nucleic acid molecules;

a) subjecting the tagged nucleic acid molecules to polymerase chain reaction (PCR) amplification;

b) generating tag associated sequence reads by sequencing the product of step (b);

c) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a mRNA transcript to a location on the cDNA library; and

d) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

Also provided is a method for obtaining from genomic material DNA methylation information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands to a polymerase reaction in the presence essentially unique primers, wherein the essentially unique primers are capable of hybridizing to the polynucleotide tail of the zero-th order strands, under conditions that promote the formation of only one complement, thereby generating tagged nucleic acid molecules;

d) separating the tagged nucleic acid molecules into a group consisting of hemi-methylated tagged nucleic acid molecules and a group consisting of unmethylated tagged nucleic acid molecules;

e) subjecting each group of step (d) to amplification by polymerase chain reaction (PCR);

f) generating tag associated sequence reads by sequencing the product of step (e);

g) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding

to a segment of the genomic material to a location on the genome; and

h) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining DNA methylation information unaffected by amplification distortion.

In an embodiment of the methods, tagging the segments to generate tagged nucleic acid molecules comprises:

a) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

b) subjecting the zero-th order derivative strands of step (a) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

c) adding a polynucleotide tail to the first order derivative strands;

d) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (b) and (d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

In an embodiment of the methods, tagging the segments to generate tagged nucleic acid molecules comprises:

a) adding a polynucleotide tail to the ends of the segments of genomic material to generate zero-th order derivative strands,

b) subjecting the zero-th order derivative strands of step (a) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (a) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

c) subjecting the product of step (b) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (a) under conditions which promote the formation of only one complement, wherein the primers of step (c) have different nucleotide sequences than the primers of step (b), and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (b) and (c) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

In an embodiment of the methods, adding a polynucleotide tail comprises the use of a terminal transferase.

In an embodiment of the methods, tagging the segments to generate tagged nucleic acid molecules comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic material.

In an embodiment of the methods, the adaptors comprising the tags are ligated to only one end of the segments of the genomic material.

In an embodiment of the methods, the tags comprise a sequence that aids PCR amplification.

In an embodiment of the methods, each tagged nucleic acid molecule comprises one tag.

In an embodiment of the methods, each tagged nucleic acid molecule comprises more than one tag.

In an embodiment of the methods, segments of the genomic material are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

In an embodiment of the methods, segments of the cDNA are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

In an embodiment of the methods, the maximum copy number of a location on a cDNA library is not less than the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library.

An embodiment of the above methods further comprises analyzing mRNA copy number.

In an embodiment of the methods, separating the tagged nucleic acid molecules into a group consisting of hemi-methylated tagged nucleic acid molecules and a group consisting of unmethylated tagged nucleic acid molecules is by cleavage with

methylation sensitive restriction enzymes, partitioning with antibodies, or partitioning with methyl-C binding proteins directed to methylated or hydroxymethylated cytosine.

In an embodiment of the methods, the tagged nucleic acid molecules are subject to hybrid capture prior to PCR or prior to sequencing.

In an embodiment of the methods, each tagged nucleic acid molecule differs at more than one nucleotide.

In an embodiment of the methods, the tag sequences further comprise a sample tag.

In an embodiment of the methods, the tagged nucleic acid molecules are pooled with a plurality of tagged nucleic acid molecules having a different sample tag prior to PCR amplification or prior to sequencing.

An embodiment of the methods further comprises deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads according to sample tag.

In an embodiment of the methods, the tagged nucleic acid molecules are generated from a single species.

In an embodiment of the methods, the tagged nucleic acid molecules are generated from a single organism.

In an embodiment of the methods, tagged nucleic acid molecules are generated from a single cell.

In an embodiment, the single cell is from a needle aspirate of suspected cancer lesions.

In an embodiment, the single cell is from a core biopsy of suspected cancer lesions.

In an embodiment of the methods, the tagged nucleic acid molecules are generated from two or more organisms.

In an embodiment, the tagged nucleic acid molecules are generated from a single cell of each organism.

In an embodiment of the methods, the single species is human.

In an embodiment of the methods, the tagged nucleic acid molecules are generated from two or more species.

In an embodiment, the tagged nucleic acid molecules are generated from a population of microbes.

A further embodiment comprises comparing the genomic copy number information obtained for different species of the population to determine the relative count of those different species in the population.

Also provided is a composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands of step (b) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

d) adding a polynucleotide tail to the first order derivative strands;

e) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (c) and (e) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

Also provided is a composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of genomic material;

b) adding a polynucleotide tail to the ends of the segments of genomic material to generate zero-th order derivative strands,

c) subjecting the zero-th order derivative strands of step (b) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (b) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

d) subjecting the product of step (c) to a polymerase reaction in the presence of primers

capable of hybridizing to the polynucleotide tail added in step (b) under conditions which promote the formation of only one complement, wherein the primers of step (d) have different nucleotide sequences than the primers of step (c), and wherein the polymerase has 3'-5' proofreading activity,

wherein the primers of at least one of steps (c) and (d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

Also provided is a composition of matter derived from a mRNA transcripts comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA; and

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, wherein each tagged nucleic acid molecule comprises

i) one cDNA from step (a) or one segment of the cDNA from step (b) and

ii) a tag.

Also provided is a composition of matter derived from mRNA transcripts comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining mRNA transcripts;

b) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

c) adding a polynucleotide tail to the first order derivative strands;

d) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (c) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (b) and d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

In an embodiment of the above compositions, adding a polynucleotide tail comprises the use of a terminal transferase.

In an embodiment of the above compositions, each tagged nucleic acid molecule comprises one tag.

In an embodiment of the above compositions, each tagged nucleic acid molecule comprises more than one tag.

In an embodiment of the above compositions, segments of the genomic material are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

In an embodiment of the above compositions, segments of the cDNA are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

In an embodiment of the above compositions, the complexity of the tagged nucleic acid molecules is reduced by hybrid capture.

In an embodiment of the above compositions, each tagged nucleic acid molecules differs at more than one nucleotide.

In an embodiment of the above compositions, the tag sequences further comprise a sample tag.

Also provided is a composition of matter comprising at least two pools of tagged nucleic acid molecules as described herein.

In an embodiment of the above compositions, the tagged nucleic acid molecules are generated from a single cell.

In an embodiment, the single cell is from a needle aspirate of suspected cancer lesions.

In an embodiment, the single cell is from a core biopsy of suspected cancer legions.

In an embodiment of the above compositions, the tagged nucleic acid molecules are generated from two or more organisms.

Also provided is a kit for determining nucleic acid copy number information unaffected by amplification distortion comprising:

a) a terminal transferase, and

b) plurality of substantially unique primers,

wherein the substantially unique primers comprise substantially unique tags, and wherein the substantially unique primers are capable of hybridizing to the polynucleotide tail of a nucleic acid molecule which has a polynucleotide tail added by the terminal transferase.

In an embodiment, the kit further comprises a DNA polymerase having 3'-5' proofreading activity.

In an embodiment of the kits, the plurality of substantially unique primers comprises 10^n primers, wherein n is an integer from 2 to 9.

In an embodiment, the kits further comprise a second terminal transferase and a primer capable of hybridizing to the polynucleotide tail of a nucleic acid molecule which has a polynucleotide tail added by the second terminal transferase.

In an embodiment of the kits, the tags are six nucleotides long.

In an embodiment of the kits, the tags are 15 nucleotides long.

In an embodiment, the kits further comprise sample tags.

In an embodiment, the sample tags are 2 nucleotides long.

In an embodiment, the sample tags are 4 nucleotides long.

In an embodiment, the sample tags comprise a sample tag set.

In an embodiment of the above kits, the substantially unique tags comprise the sample tags.

Terms

For the purpose of this invention, different words and phrases are defined as follows:

As used herein, the term "adaptor" refers to an oligonucleotide or nucleic acid fragment or segment that can be ligated to a nucleic acid molecule of interest. For the purposes of this invention adaptors may, as options, comprise primer binding sites, recognition sites for endonucleases, common sequences, promoters, tag sequences, and sample tag sequences. Preferably, adaptors are positioned to be located on both sides (flanking) a particular nucleic acid molecule of interest. In accordance with the invention, adaptors may be added to nucleic acid molecules of interest by standard recombinant techniques (e.g. restriction digest and ligation). For example, adaptors may be added to a population of linear molecules, (e.g. a genomic DNA which has been cleaved or digested) to form a population of linear molecules containing adaptors at one and preferably both termini of all or a substantial portion of molecules. The adaptor may be entirely or substantially double stranded or entirely single stranded. A double stranded adaptor may comprise two oligonucleotides that are at least partially complementary. The adaptor may be phosphorylated or unphosphorylated on one or both strands. Adaptors may be used for DNA sequencing. Adaptors may also incorporate modified nucleotides that modify the properties of the adaptor sequence. For example, methylated cytosines may be substituted for cytosines. In an embodiment of this invention the adaptors ligated to genomic DNA to enable cluster generation on the sequencer contain cytosines which were all methylated. This modification protects such adaptors from bisulfite conversion, and is taken into account in the downstream applications and analysis of this invention.

As used herein, the term "amplifying" refers to the process of synthesizing nucleic acid molecules that are complementary to one or both strands of a template nucleic acid. Amplifying a nucleic acid molecule typically includes denaturing the

template nucleic acid, annealing primers to the template nucleic acid at a temperature that is below the melting temperatures of the primers, and enzymatically elongating from the primers to generate an amplification product. The denaturing, annealing and elongating steps each can be performed once. Generally, however, the denaturing, annealing and elongating steps are performed multiple times (e.g., polymerase chain reaction (PCR)) such that the amount of amplification product is increasing, often times exponentially, although exponential amplification is not required by the present methods. Amplification typically requires the presence of deoxyribonucleoside triphosphates, a DNA polymerase enzyme and an appropriate buffer and/or co-factors for optimal activity of the polymerase enzyme. The term "amplification product" refers to the nucleic acid sequences, which are produced from the amplifying process as defined herein.

As used herein, the term "amplification distortion" refers to non-uniform amplification of template nucleic acid molecules.

As used herein, the term "bisulfite-treatment" refers to the treatment of nucleic acid with a reagent used for the bisulfite conversion of cytosine to uracil. Examples of bisulfite conversion reagents include but are not limited to treatment with a bisulfite, a disulfite or a hydrogensulfite compound.

As used herein, the term "bisulfite-converted material" refers to a nucleic acid that has been contacted with bisulfite ion in an amount appropriate for bisulfite conversion protocols known in the art. Thus, the term "bisulfite-converted material" includes nucleic acids that have been contacted with, for example, magnesium bisulfite or sodium bisulfite, prior to treatment with base.

As used herein, the term "sequence capture" or "hybrid capture" refers to a process of hybridizing a "capture probe" to a nucleic acid having a sequence which is complementary to the sequence of the capture probe. A capture probe may be immobilized to a substrate on a solid phase microarray, wherein "substrate" refers to short nucleic acid sequences which are known. Their location on the solid phase microarray may or may not be predetermined. The capture probe comprising a "sequence complementary to the substrate" may be immobilized to the solid phase microarray by hybridizing to its complementary "substrate sequence."

As used herein, the term "clamp" refers to a nucleotide sequence of CC, CG, GC or GG which may be located at the first two nucleotide positions at the 5' end of a primer, tag, or sample tag, or the last two nucleotide positions at the 3' end of a primer, tag, or sample tag.

As used herein, the term "comparable number of locations" means that a first genomic region has a number of locations on the genome which is within 25% of the number of locations of a second genomic region. In an embodiment, the first genomic region which has a comparable number of locations to the second genomic region has a number of locations which is within 20%, 15%, 10%, 5%, 1%, or exactly the same as the number of locations of the second genomic location.

As used herein, a subsequence of a tag associated sequence read is "corresponding to" a tag when the subsequence is identical to the nucleotide sequence of the tag.

As used herein, a subsequence of a tag associated sequence read is "corresponding to" a species of a nucleic acid molecule when the subsequence is substantially identical to or substantially complementary to at least about 10, 12, 14, 16, 18, 20, or more nucleotides of the sequence of the species of

nucleic acid molecule. In an embodiment, the subsequence is identical to or fully complementary to at least about 10, 12, 14, 16, 18, 20, or more nucleotides of the sequence of the species of nucleic acid molecule.

The term "CpG site" refers to the CpG dinucleotide.

As used here, the term "CpG island" refers to a region of DNA with a high G+C content and a high frequency of CpG dinucleotides relative to the bulk genome, as further defined at the UCSC Genome Bioinformatics Site at genome.ucsc.edu/index.html?org=Human&db=hg19&hgsid=171216665, and in particular, by annotation in the USCS Genome Bioinformatics Site database (CpG Islands Track or Table: cpgIslandExt).

As used herein, the term "fully complementary" refers to the reverse complement of a nucleic acid sequence.

As used herein, the term "library" refers to a collection of nucleic acid molecules (circular or linear). In one preferred embodiment, a library is representative of all of the DNA content of an organism (such a library is referred to as a "genomic" library), or a set of nucleic acid molecules representative of all of the expressed genes (such a library is referred to as a cDNA library) in a cell, tissue, organ or organism. A library may also comprise random sequences made by de novo synthesis, mutagenesis of one or more sequences and the like. A library may be contained in one vector.

As used herein, the term "mapping" refers to identifying a location on a genome or cDNA library that has a sequence which is substantially identical to or substantially fully complementary to the subsequence of a tag associated sequence read corresponding to a species of nucleic acid molecule, and assigning the tag associated sequence read or the tagged nucleic acid molecule generating the tag associated sequence

read to the location. The nucleic acid molecule may be, but is not limited to the following: a segment of genomic material, a cDNA, a mRNA, or a segment of a cDNA.

As used herein, the term "methylation" refers to the covalent attachment of a methyl group at the C5-position of the nucleotide base cytosine within the CpG dinucleotides of genomic region of interest. The term "methylation state" or refers to the presence or absence of 5-methyl-cytosine ("5-Me") at one or a plurality of CpG dinucleotides within a DNA sequence. A methylation site is a sequence of contiguous linked nucleotides that is recognized and methylated by a sequence specific methylase. A methylase is an enzyme that methylates (i. e., covalently attaches a methyl group) one or more nucleotides at a methylation site.

As used herein, the term "mRNA transcript" refers to the nucleic acid molecule resulting from transcription of DNA.

As used herein, the term "pool" means to combine one plurality of nucleic acid molecules with at least one other plurality of nucleic acid molecules. In an embodiment, tagged nucleic acid molecules are pooled. Pooling may be done after any step in which tagged nucleic acid molecules are generated. In an embodiment, pooling is done prior to PCR and/or prior to sequencing. In an embodiment, tagged nucleic acid molecules generated from a single cell are pooled with tagged nucleic acid molecules generated from a different single cell.

As used herein, the term "probe array" refers to an array of N different nucleic acid molecules deposited on a reaction substrate which serve to interrogate mixtures of target molecules or multiple sites on a single target molecule administered to the surface of the array.

As used herein, the term "read" or "sequence read" refers to the nucleotide or base sequence information of a nucleic acid that has been generated by any sequencing method. A read therefore corresponds to the sequence information obtained from one strand of a nucleic acid fragment. For example, a DNA fragment where sequence has been generated from one strand in a single reaction will result in a single read. However, multiple reads for the same DNA strand can be generated where multiple copies of that DNA fragment exist in a sequencing project or where the strand has been sequenced multiple times. A read therefore corresponds to the purine or pyrimidine base calls or sequence determinations of a particular sequencing reaction.

As used herein, the term "reference genome" refers to a genome of the same species as that being analyzed for which genome the sequence information is known.

As used herein, the term "region of the genome" refers to a continuous genomic sequence comprising multiple discrete locations.

As used herein, the term "representation of DNA" or "representation of a genome" refers to a sampling of the DNA or genome produced by a restriction endonuclease digestion of genomic or other DNA, followed by linkage of adaptors and then amplification with primers complementary to the adaptors.

As used herein, the term "sample tag" refers to a nucleic acid having a sequence no greater than 1000 nucleotides and no less than two that may be covalently attached to each member of a plurality of tagged nucleic acid molecules or tagged reagent molecules. A "sample tag" may comprise part of a "tag."

As used herein, the term "sample tag set" refers to a plurality of unique sample tags of the same length, wherein the nucleotide sequence of each unique sample tag differs from

the nucleotide sequence of any other unique sample tag in the sample tag set at two or more corresponding positions in the sequence. A unique sample tag selected from the sample tag set may be covalently attached to each member of a unique sample consisting of a plurality of tagged nucleic acid molecules in order to identify a tagged nucleic acid molecule as a member of that unique sample.

As used herein, the term "segments of cDNA," refers to the nucleic acid molecules resulting from fragmentation of cDNA.

As used herein, the term "segments of genomic material" refers to the nucleic acid molecules resulting from fragmentation of genomic DNA.

A nucleic acid molecule which contains a sequence of nucleotides identical to a segment of a cDNA or a segment of genomic material comprises a "segment of cDNA" or a "segment of genomic material," respectively.

As used herein, the "sequence complexity" or "complexity" with regards to a population of polynucleotides refers to the number of different species of polynucleotides present in the population.

As used herein, "substantially identical" or "substantially fully complementary" sequences have at least about 80% sequence identity or complementarity, respectively, to a nucleotide sequence. Substantially identical sequences or substantially fully complementary sequences may have at least about 85%, 90%, 95% or 100% sequence identity or complementarity, respectively.

As used herein, the term "substantially unique primers" refers to a plurality of primers, wherein each primer comprises a tag, and wherein at least 50% of the tags of the plurality of

primers are unique. Preferably, the tags are at least 60%, 70%, 80%, 90%, or 100% unique tags.

As used herein, the term "substantially unique tags" refers to tags in a plurality of tags, wherein at least 50% of the tags of the plurality are unique to the plurality of tags. Preferably, substantially unique tags will be at least 60%, 70%, 80%, 90%, or 100% unique tags.

As used herein, the term "tag" refers to a nucleic acid having a sequence no greater than 1000 nucleotides and no less than two that may be covalently attached to a nucleic acid molecule or reagent molecule. A tag may comprise a part of an adaptor or a primer.

As used herein, a "tagged nucleic acid molecule" refers to a nucleic acid molecule which is covalently attached to a "tag."

Where a range of values is provided, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limit of that range, and any other stated or intervening value in that stated range, is encompassed within the invention. The upper and lower limits of these smaller ranges may independently be included in the smaller ranges, and are also encompassed within the invention, subject to any specifically excluded limit in the stated range. Where the stated range includes one or both of the limits, ranges excluding either or both of those included limits are also included in the invention.

All combinations of the embodiments of the invention described herein are within the scope of the invention unless clearly indicated otherwise.

Single Cell Copy Number Profiling by 'Sequence Counting' and Sample Tagging.

High throughput sequencers are in effect single molecule sequencers. Because the human genome is sequenced and assembled, a single molecule sequencer is also a single molecule identifier. Therefore, sequencing is also counting, as illustrated in Figure 4 which illustrates how low coverage sequencing can yield an estimate of copy number which is comparable to array hybridization. The distinction for this is that 'sequence counting' can be done on single cells, and, using the multiplex system disclosed herein, relatively inexpensively, in effect converting the high throughput sequencer into a detector of single cells with abnormal genomes. To the sequencing technology two additional features were added: limiting dilution and sample tagging. For single-cell sequencing, the single cells or the nuclei of the single cells are first deposited into separate wells or are kept physically separated by other means, i.e. a gel. The single cells or the nuclei of cells can be deposited in a well by a cell sorter. Alternatively a collection of cells or nuclei can be diluted into a fluid that is then made semi-solid. A unique sample tag is prepared for each well for sequencing, with a separate sample tag per well. After this step, the wells are pooled and sequenced. The sample tag is used to deconvolute the information, allowing a copy number profile to be made for each individual cell.

Protocol: Isolate single tumor cells or single nuclei of the tumor cell, tag and optionally sample tag the DNA of each cell or nuclei, and amplify the tagged DNA to microgram quantities to generate tagged libraries for Solexa sequencing. The isolation of single tumor cells will occur by Fluoresent Activated Cell Sorting by total genomic DNA content, to deposit single nuclei into individual wells.

Alternatively, tumor cells can be isolated by FACS sorting with fluorescent antibodies against breast epithelium markers,

including cytokeratin 8, 18 or 19. Cells are lysed *in situ* and the genome is fragmented by heating. Libraries are constructed from the fragmented genomic DNA. Preliminary experiments were conducted using high-resolution ROMA microarray to quantify whole-genome copy number on DNA amplified using WGA from single SK-BR-3 cells (Figure 5). These experiments demonstrate the feasibility and efficiency of amplifying 6 pg of DNA into microgram quantities to determine copy number by microarray methods, which suggests that the DNA is also suitable for determining copy number by sequencing.

Since sequencing whole tumor genomes from single cells is costly and time consuming, two methods are proposed to stratify the genome and sequence for copy number: Representation Sequencing and Bin Sequencing.

Representation Sequencing

Representation sequencing involves restriction digesting genomic DNA into fragments and sequencing only the stratified population of restriction fragments to determine copy number. This method has the advantage of generating precise restriction fragments that can be reliably prepared as libraries from different individual tumor cells. In this protocol genomic DNA is amplified from a single tumor cell, digested with the DpnII, a second digestion is preformed with a AluI enzyme to remove highly repetitive "Alu SINE elements," sequencing adaptors (Illumina GA, Solexa) with a unique 4 nucleotide sample tag are ligated and the ligated fragments are PCR amplified to generate a sequencing library. *In silico* modeling predicts that the double digest will increase the fraction of unique sequencing reads to 75% from the 55% normally seen in unselected human genome sequencing. The sample tagged DNA libraries are then pooled, and applied to the Solexa flow-cells for cluster amplification and single

read sequencing. Single-end reads are used because they are faster and cheaper than paired-end reads and provide all of the information necessary for 'sequence counting'. Mapping algorithms then align each sequence read with a high quality score (with a read length in excess of 35bp) and an unambiguous sample tag to a specific location on the DpnII restriction map of the human genome. The result is a count of reads at places in the genome, similar to the process demonstrated in Figure 4, except that the data is segregated by sample tag, so that each well receives a copy number profile.

Bin Sequencing

It is clear that sequencing of random size-selected fragment from the genome is a viable alternative. In this method, the DNA from single tumor cells is amplified to generate fragments from 200-2000bp. Blunt-end cloning is then used to ligate adaptors to these fragments, followed by PCR amplification to generate libraries (standard Solexa protocol for single read genomic DNA). However, in addition to this protocol adaptor sequences with 4 nucleotide sample tags are ligated to generate unique libraries from each tumor cell genome. The sample tagged DNA libraries are then pooled, and applied to the Solexa flow-cells for cluster amplification and single read sequencing. The 35bp sequences are then mapped to specific regions of the human genome and deconvoluted using the unique sample tag to determine which sequence reads originated from which tumor cells. The crucial step in this procedure that is different from representation methods is to artificially divide the human genome into "bins" of several kilobases. Break analysis in sequence count data has another degree of freedom compared to what is practiced for array data, namely that the boundaries of the bins can be adjusted so as to minimize variance. Each bin contains a number of

random genomic sequence reads from which a mean value is computed to determine genomic copy number at low sequence coverage.

The particular use of the WGA method does not depend on absolutely uniform amplification across the genome. Useful data can be obtained even if only half of the potential fragments from a single cell were amplified in a given reaction. Additionally, redundancy of data gained from segmentation benefits in identifying breakpoints.

Representation Sequencing vs. Bin Sequencing

A comparison of the two methods reveals the number of samples that can be profiled per sequencing run. Of the 3,149,324 Dpn II fragments in size range 200-800 (size optimal for sequencing) there are 618,629 not cut with AluI and thus available to be amplified. Approximately 75% of the fragment ends, or about 450,000, yield unique mappings and thus provide locations for counting. The overall process is presented as a flowchart in Figure 7.

Assuming 15 million quality reads per Solexa sequencing lane, random sequencing yields about 55% or 8.25 million unique reads per lane. The DpnII/AluI representation should increase the number of unique reads to 11.25 million. Modeling a single flow cell lane with 100kb bins distributed across the genome, the projected results are as follows:

Table 1

# samples	avg reads/bin/sample (random)	avg.reads/bin/sample (Dpn/Alu)
1 sample per lane	267	363
2 samples per lane	133	181
12 samples per lane	22	30

The number of samples (individual cells) to be sequenced per lane, is between 4 and ten. With 8 lanes on a flowcell, 30-80 samples could be profiled at the level of Figure 4 relatively inexpensively.

Finally, the whole genomic copy number (generated by sequencing) will be determined for multiple, individual tumor cells in a single solid tumor. The data will be segmented into sections that minimize variance of copy number, subject to Kolmogorov-Smirnov significance tests, to generate individual tumor copy number profiles. As stated above, an additional degree of freedom, which our algorithms do not yet incorporate, is selecting the "boundary" of the bins. The single cell tumor genome profiles will be compared to study tumor heterogeneity in single solid tumors which may elucidate the progression of genomic events during tumorigenesis.

Varietal counting of nucleic acids

Measuring the absolute or relative concentration of nucleic acid sequences in a sample often suffers from severe stochastic distortions due to differential amplifications or losses during processing and manipulation of the nucleic acids. There is never more information available than the number of molecules of the starting material in the sample

itself. The method described herein is designed to capture the information at the earliest stage, in a form that is then resistant to distortion by further amplification.

Input nucleic acid molecules (0-th order derivatives), or first round copies of them (1-st order derivatives), present in the sample, can be randomly tagged using a large choice of distinct tags. Every tagged molecule becomes essentially unique, the combination of the information in the tag and the information in the nucleic acid, which can later be read by sequencing. The combination of a tag with a 0-th order, 1-st order, or subsequent N-th order nucleic acid molecule is called a "tagged nucleic acid molecule." After amplification or selection or other processing, the number of different tags is counted.

Within a pool of tagged nucleic acid molecules, each tagged nucleic acid molecule is likely unique in the pool when a sufficiently large number of distinct tags is used. Then in subsequent amplifications these unique tagged nucleic acid molecules are amplified to facilitate detection, but no new species of tagged nucleic acid molecules are created. Thus, distorted counts are not created provided that the number of different tagged nucleic acid molecules is counted.

In the disclosed method, each sequence read generated from a tagged nucleic acid molecule comprises two parts, or subsequences. The first part corresponds to the tag, which identifies the tag (and the sample tag if samples are pooled). The second part corresponds to a nucleic acid in the sample which is mapped to a location in the genome (or to a transcript if RNA molecules are being counted). After deconvoluting by sample tag, the number of different tagged nucleic acid molecules mapped to each location is counted. There are two separate methods of counting, and each can be combined for greater accuracy and determination of confidence.

The first counting method is called "maximum tagged nucleic acid molecule number." The number of different tagged nucleic acid molecules at a given location cannot exceed the true (absolute) copy number. Therefore, for a given region comprising multiple locations, the maximum copy number of the region is not less than the maximum number of different tagged nucleic acid molecules mapped to any location in that region. The maximum number of different tagged nucleic acid molecules centered about a moving window of a fixed number of locations can be taken as the measurement of true copy number of the window. This will provide at worst an underestimate of the maximum true copy number for that window, and never an overestimate. The set of maximum number of different tagged nucleic acid molecules is one type of copy number profile.

The counting method described above is most accurate when the processing efficiency is excellent and the number of molecules mapped to each location is less than the number of tags. Under such conditions virtually each molecule in the sample is tagged and counted, and the true count can be derived. But there is a second method.

The second counting method is called "total different tagged nucleic acid molecules," and is useful, for example, when tagging efficiency is low. The total number of different tagged nucleic acid molecules within a region of a fixed number of locations will be, within a statistical counting error, a monotonic function of true copy number, regardless of the efficiency of tagging. Low efficiency of tagging merely increases the statistical error. The relative copy number of a region can be estimated by comparing the total number of different tagged nucleic acid molecules mapped to the region to, for example, the mean value of tagged nucleic acid molecules mapped to other regions of the genome which have a comparable number of locations.

If the number of molecules at a location on a genome or cDNA library exceeds the number of tags, and tagging is too efficient, copy number information can not be recovered. Rather, the total number of tags at each location would merely be measured. There are at least three ways to remedy this condition: 1) diminish the efficiency of the first step of the process, for example by reducing the reaction time; 2) reduce the amount of sample; and 3) increase the total number of tags. For example, if the length of the tag were N nucleotides, there are 4^N possible tags. For N = 15, there are approximately 10^9 tags, easily in excess of the number of molecules per location.

The collection of reads lend themselves to modeling the power of inference. For each location, there exists the number of tagged nucleic acid molecules observed, and for each species of tagged nucleic acid molecule, the number of times it was observed. With a good estimate of the input, the number of genomes, critical parameters can be accurately deduced because the vast majority of the copy numbers are two per genome. The limiting case of single cell analysis lends itself to the most accurate modeling, with the proportion of null reads enabling the inference of the probability, theta, that a single molecule at a location is detected. Theta is a function of the efficiency of tagging, the efficiency of amplification and processing, and the read depth.

Using the language invented of the WGA reaction, in the specific protocol described here, all first order derivative DNA molecules (i.e., first copies) are tagged, with no new tags introduced when subsequent order derivatives are created. This enables the "counting" of original molecules, alleles in the case of genome analysis. This method is referred to herein as "varietal counting." As long as sequence depth is sufficient, subsequent amplification or methods of trapping

that enrich for regions of interest, or the combination, cannot distort the count. Pooling from multiple cells becomes a natural extension of this method, which improves efficiency, increases throughput and decreases costs.

Tags

The tags are nucleotide sequences with constant portions and variable portions. The length of the constant portion and the length of the variable portion may be the same or different. The length of the tags within a plurality of tags is typically, but not necessarily, the same for all tags. In an embodiment, the total length of a tag is less than 100 nucleotides.

The constant portions are used to aid in the tagging of nucleic acid molecules. For example, the constant portion may contain a nucleotide sequence which allows hybridization to a poly-N tail added by a terminal transferase. The constant portion may also be used to aid in manipulation of the tagged nucleic acid molecules. For example, the constant portion may contain a sequence which is a primer binding site useful for PCR amplification. The constant portion may also be used to identify a sample if there are multiple samples being processed in parallel, i.e., the constant portion may contain a sample tag.

The sequence of individual tags does not need to be known prior to generating tagged nucleic acid molecules. A plurality of tags may have variable portions whose sequences are elucidated for the first time during sequencing. The variable portion of a tag may be a sequence of nucleotides where each nucleotide is individually one of A, T, C, or G, or one of any two of A, T, C, and G, or one of any three of A, T, C, and G, i.e. the variable portion of a tag may consist of only two or three species of nucleotides. Tags may be designed such that

the variable portion has a random sequence. Tags may be designed such that the variable portions are constructed from a set of dinucleotides or trinucleotides.

Tags may be associated with nucleic acid molecules by using, singly or in combination, ligation, nick-ligation, terminal transferase, hybridization, priming, and the like. For example, nucleic acid molecules may be cut with a restriction endonuclease(s), treated with a terminal transferase to add a poly-N tail, hybridized to a tag molecule, and then nick ligated to form a tagged nucleic acid molecule. Specific methods of generating tagged nucleic acid molecules are discussed in detail below.

Tagging with the aid of a terminal transferase

Figure 8 illustrates one embodiment of this method, where the N_{10} designates an oligonucleotide having a tag component and a sample tag component. The first six nucleotides (4096 possible sequences), randomly generated, provide the tag component, and the last four nucleotides (256 choices) are chosen to encode the micro-well, and, hence, a sample tag. The lengths of the tag and the sample tag can be changed to suit needs, and are not limited to a total of ten nucleotides. The lengths are chosen for illustration. The sample tag allows pooling, with deconvolution occurring afterwards. The combination of a tag with a nucleic acid molecule from the input sample provides an essentially unique tagged nucleic acid molecule for the first derivative strand, that can only with very low probability be replicated by chance from another like molecule (a chance of roughly 1/N where N is the number of available tags). In the end, multiple copies of sequence-identical molecules in the input sample are each individually counted. In applications to mRNA counting, the length of the tag component can be increased to accommodate possibly thousands of transcripts from the same gene.

Although there are multiple embodiments of the basic method besides the one illustrated in Figure 8, the embodiment illustrated in Figure 8 has several useful design features. First, it avoids the use of ligases (an alternate method for associating a tag with a nucleic acid molecule), which has been found to be an inefficient reaction. Instead, this embodiment uses terminal transferase (TTase), which is a robust enzyme from which high efficiency is expected. Second, once the tag and sample tag are incorporated into the first derivative DNA strand, micro-wells can be pooled and the subsequent steps carried out more efficiently in larger volumes with larger numbers of molecules, possibly with the addition of carrier nucleic acids. Because of a clean-up step, removing free adaptor, no more unique first order derivative strands with tags can be created, and second order strands can be arithmetically expanded if found to be useful. Third, the method extends immediately from DNA to mRNA. Rather than cleaving and adding a polynucleotide tail with terminal transferase, the naturally occurring polyA tail of mRNAs can be utilized. Fourth, it is easy to see how to combine analysis of both RNA and DNA from the same cell with this schema. Instead of tailing DNA with A, it can be tailed with, for example, C, thus distinguishing the DNA from the RNA. DNA and RNA derivatives can either be read together in the same sequencing runs, or separately by amplification using their differing PCR primer adaptors. Fifth, by using a longer sample tag, sequences belonging to a particular cell can be preferentially amplified for deeper study when warranted.

This method can be applied to assessing DNA methylation. After the first order derivative strands are made, the duplex DNA can be pooled from multiple samples. These molecules will be hemi-methylated, and still retain epigenetic marks. Therefore they can then be separated into methylated and unmethylated samples either by cleavage with methylation sensitive

restriction enzymes, or by partitioning with antibodies or methyl-C binding proteins directed to methylated (or hydroxyl methylated) cytosine. Error rates will be measured based on expectations from regions of known methylation state.

Tagging by ligation

In another embodiment of the invention, adaptors with tag(s) can be ligated to sample DNA or RNA that has been converted to DNA. In the second step these molecules can be pooled (or not) and amplified for sequencing. In the third step the numbers of each different tagged nucleic acid molecule of each locus (or mappable sequence read) are compiled, giving statistics from which copy number can be inferred.

In another embodiment, to measure RNA, RNA is first converted to DNA using methods that produce a single double stranded molecule for each RNA in the sample. (For example, prime with oligo dT and reverse transcriptase, followed with random priming and Klenow polymerase.) The procedures for analyzing DNA can then be followed.

In another embodiment, the sample DNA can be sheared and the ends prepared for ligation. However, there are three major advantages to cleaving with one or more restriction endonucleases (RE). The first is that the expected number of tagged nucleic acid molecules at each location can be precisely predicted from the starting number of cells in the sample, or conversely the number of cells in the sample inferred from the expected number of tagged nucleic acid molecules per location. The second is that the expected complexity of the product is reduced, enabling copy number determination from fewer sequence reads. The third are certain advantages in the flexibility and functionality that can be built into the tags.

In an embodiment, sample nucleic acid is cleaved with a specific restriction enzyme and sample molecules are ligated to tagged (and sample tagged if appropriate) oligonucleotide adaptors which contain sequences that allow PCR amplification. There are various designs for these adaptors, and also alternatives to PCR (such as WGA), based on the functionalities built into the tags. Some of these additional functionalities are discussed below.

The ligated molecules now are essentially unique, the ligated molecules each being the combination of a substantially unique tag and DNA sequence to which they were ligated. Further amplification may distort the yields of molecules, but will not create new tags (although some may be lost by statistical processes which are readily modeled during analysis).

Through sequence error during processing, the tag may very occasionally mutate, creating the appearance of a new tagged nucleic acid molecule. But if the set of tags are sufficiently large, the tagged nucleic acid molecules will generally differ at more than one nucleotide, and so tagged nucleic acid molecules that differ by a single nucleotide can (optionally) be ignored when counting.

For single cells, individual cells are loaded at most one per microwell. The nucleic acid is then prepared in wells, and the molecules tagged. Once tagged they can be pooled for further processing, which greatly improves the uniformity and efficiency of DNA amplification methods.

In yet another embodiment, DNA is cut, and adaptors are ligated to both ends of each fragment, enabling them to be amplifiable by PCR. To avoid adaptor-adaptor self ligation, double stranded adaptors without 5′ phosphorylation can be used, so an adaptor is ligated on one strand only at each end of the cleaved sample DNA. Then the molecules are pooled, and

treated as for representations: the shorter single strand adaptor is melted off at elevated temperature but well below the Tm for the sample duplex, the ends of the double stranded DNA filled in with Klenow polymerase, unligated adaptors removed, and then the sample is PCR amplified.

In the preceding embodiment, adaptors are used not only for tagging but also for amplification. Adaptors are for identifying each molecule individually. They can also be used for amplification, as described above, or for other uses (see below). Because in the above embodiment adaptor ligation at both ends of sample fragments is required for amplification, the yield may be diminished. To improve yield, the method can work with molecules that have an adaptor at one end only.

In one embodiment, the reactions are performed as before, but tags do not contain sequences enabling PCR. Prior to PCR, but after an optional pooling, fresh adaptors (different composition and without tags) are ligated so that molecules with only a single tag adaptor can be amplified and eventually counted. If the original tag has a selection sequence, molecules with the tag adaptor before sequencing can be optionally enriched by hybrid selection.

In another embodiment, initial adaptors are designed with a phosphorylated 5′ end and a 3′ dideoxy end, so that the initial ligation occurs to the opposite strand as described in the embodiment above. (Instead of using an RE with a 5′ overhang, one which leaves a 3′ overhang may be preferred.) After removal of unligated adaptors, there follows several rounds of arithmetic amplification, followed by tailing molecules with dT using terminal transferase, and amplification of the processed sample DNA by PCR.

In another embodiment, DNA from sample is labeled as before, pooled if required, and unligated adaptors removed from the

reaction. The reaction is subject to whole genome amplification (WGA), and the product of WGA is used to make sequencing libraries. For this embodiment, the initial adaptors may also contain: (A) cleavage sites for ease of making sequencing libraries; and (B) a sequence large enough for optional enrichment by hybrid selection prior to making libraries for sequencing.

In the embodiments just described, it is assumed that the tag is double stranded. The production of a great variety of double stranded molecules is not completely straightforward. In one embodiment, single strand DNA is synthesized as type I and type II. Type II has three sets of functionalities A, B, C, and D. Functionalities B contain the tag sequence, C the adhesive ends with a RE site, and D the buffer. The B will contain some length of runs with random sequence, for example 10 nucleotides with a total complexity of 4^{10}, or about 10^6 possible sequences. Type I oligo will be complementary to type II at functionality A, the two joined and annealed, and then extended with polymerase to create a double stranded molecule. After cleavage with the RE site, the resulting double stranded molecule is the tag used for the subsequent tagging of the sample.

Additional methods for producing single and double stranded oligonucleotides suitable for use as tags are known in the art, for example, in U.S. Patent Nos. 5,639,603 and 7,622,281, U.S. Patent Application Publication No. 2006/0073506 and Parameswaran et al. (2007) (7,5,8,4).

Nick-ligation

In this embodiment of the invention, 3'OH ends of nucleic acid molecules, e.g., segments of genomic material or mRNA transcripts, are first extended with terminal transferase, adding a poly-N tail. Primer adaptors are then annealed to the

poly-N tails and the resulting duplex with a nick is sealed with DNA ligase. This series of reactions is much more efficient than a single step double stranded ligation. The resulting product is amplified by PCR. This method has been successfully used with as little as 1ng of DNA. Tagged nucleic acid molecules generated using the nick-ligation method can be generated from a single cell DNA, and then pooled prior to PCR. Pooling from 100 cells yields the DNA concentration needed for successful amplification. Sample tags can be added in the nick ligation step, and this is readily achieved (see Figure 9).

Technology for inexpensive assay of regional markers

The above description is of methodology designed to measure copy number and mutational loads throughout much of the genome in single cells. But the application of discoveries, once made, of predictive markers for outcome or therapeutic response will not require whole genome information. Our present outcome markers are in fact copy number markers at very specific loci. Essentially the same methodology as disclosed hereinabove can be employed to make these assessments very affordable. Consider, for example, the case where it is desirable to assess a large study for the predicative power of a set of N markers, and these markers are copy number markers that can be assessed by varietal counting. In principle, sample coding, pooling, and parallel capture and sequencing can be used to assay a large number of samples in a single sequencing lane. The major difference is that we are not restricted to minute quantities of DNA, so WGA is not needed, and assays will be more robust. We have shown in our autism work that excellent copy number data can be obtained from read depth after capture. Thus we can use direct library preparation with just sample tags. The target area for capture is likely to be different and smaller than the exome. Thus we

can pool more samples per sequencing lane, and the sequencing costs for assaying hundreds of markers in hundreds of samples can be reduced. Of course, there are costs associated with the development and testing of the capture reagents, and the cost of the capture itself, and these are not negligible, but if a test were to come into widespread clinical use, it would be inexpensive, and this is one of our major objectives. There is a clear alternative, namely creating a bank of region specific PCR primers with tags and sample tags. The tags would be necessary because random unique ends created by shearing would not be available for counting based methods. This alternative plan could be less expensive and more robust than capture (see Figure 10).

Single cell RNA measurements

Several methods have now been published whereby researchers have profiled the RNA content of single cells by sequencing (Tang et al., 2009). This is potentially a very powerful method for analysis of tumor cells. A varietal counting method could be very useful for RNA profiling from single cells, and for the same reasons that it is useful with DNA. When variety is not introduced by nucleic acid processing varietal counting provides a way to avoid the distortions created by PCR. RNA and DNA can be analyzed from the same cells. The DNA analysis enables us to cluster cells into stromal and tumor subpopulations, and this in turn facilitates the interpretation of the RNA profiling. As an example of utility, suppose one is interested in searching thousands of circulating cells, or cells from a biopsy, or from the margins of a tumor, for rare tumor cells. DNA copy number can facilitate its identification, and the RNA from that cell can then be reliably used to infer additional properties, such as the tissue of origin. This combination is especially powerful

for the early detection of the new incidence or recurrence of cancer.

Examples

Methylation sequencing

Currently, high throughput sequencing requires physical purification of sub-genomic regions of interest in order to obtain high coverage of those regions. The method of sequence capture has been applied to examine methylation in large sets of CpG islands at the sequence level. Methylation sequencing is based on the conversion of C residues to T using bisulfite reagents, with methylated C's detected as those protected from C to T conversion. This means either that sequence capture must take place before bisulfite treatment, which requires more starting DNA than is available for patient specimens, or that sequence capture must be designed to accommodate the C to T conversion. The bisulfite based sequencing method used herein adopts the latter and so is suitable for high throughput single molecule sequencing by either the Illumina or Roche 454 methods, using as little as 100 ng of input DNA. The sequencing method depends on hybrid capture of bisulfite treated DNA followed by sequencing through the Illumina Solexa sequencer and mapping back to the genome using an algorithm developed Dr. Andrew Smith (19). It was observed that CpG islands can exhibit multiple different levels of methylation, from imprinted (allelic) to randomly partially methylated, to completely methylated or unmethylated. In particular, one observation that is important for this invention is that many islands switch from completely unmethylated in normal tissues to virtually completely methylated in matched tumor tissue ("DNA-methylation switch domains"). Figure 3 shows three examples of the simplest cases in normal cells versus the breast cancer cell line MDA-MB-231: stably unmethylated (A and B); the ALX gene, switched from unmethylated to completely

methylated (C and D); and a case, SSTR4, where only a segment of the island is switched (E and F). This complete switch makes it possible to detect rare tumor cell DNA in bodily fluids or other cell populations using nested PCR of bisulfite treated DNA and opens up the possibility of a creating a method for the early detection of recurrence in the form of circulating metastatic cells.

Comparative genomic hybridization (CGH) by high throughput sequencing

Until now the most economical method for high resolution genomic analysis has employed some form of microarray. The rapid rise of 'next generation' DNA sequencing is changing that landscape. Both the Illumina (Solexa) and Roche 454 instruments can provide millions of individual sequence reads randomly distributed across any input DNA (Craig et al., 2008). The depth of coverage of these technologies depends on the total length of the input DNA, from whole genome (large breadth, lower depth) to narrowly focused regions achieved by hybrid capture of specific sequences (narrow breadth, higher depth) (Albert et al., 2007; Hodges et al., 2007; Okou et al., 2007). Converting sequencing reads to copy number analysis is a matter of mapping reads and then put them into 'bins' of a certain size, depending on the purpose, counting up the numbers in the bins and performing appropriate statistical analysis to define copy number change breakpoints.

Sequence counting was applied to the degraded formalin-fixed paraffin-embedded (FFPE) DNA that was available from retrospective clinical trials with very encouraging results. Figure 4 displays a comparison of microarray and sequence counting results in a region around Her2 on chromosome 17 for a 10 year old paraffin-embedded clinical trial sample JZ33. The gray and orange points are (respectively) the raw ratio data for 244K Agilent microarray and one lane of Solexa

sequencing reads divided into 30K bins. Note that the sequencing (orange) track shows significantly less noise. The green and blue lines are the result of segmenting each data type. The segments are generally concordant, but the sequence counting method reveals a somewhat more detailed pattern. Another feature of note from Figure 4 is the robustness of our segmenting program, irrespective of the variation of noise in the two sets of raw data.

The efficacy of sequence counting on FFPE DNA and the rapidly decreasing cost of DNA sequencing in general provides an opportunity to convert our previous microarray methods to a highly flexible and scalable platform for breast cancer genome diagnostics. Both the breadth of DNA to be sequenced and the size and location (and therefore resolution) of individual bins can be easily adjusted to fit the particular loci to be assayed.

Single cell microarray and sequencing

A representation of DNA amplified from single cells was used for genomic profiling by 'sequence counting'. As a preliminary step in developing that technology, DNA was amplified from batches of 1, 10 and 100 cells of the cell line SKBR3 using the Whole Genome Amplification (WGA) Kit from Sigma-Aldrich and compared these amplified representations to equivalent microarrays on the same format done with a standard SKBR3 DNA prep. Individual cells were picked from adherent culture dishes using a cell transfer micropipette. After random WGA1 amplification the DNA preps were digested with DpnII and AluI, adaptors added and only DpnII fragments without AluI sites were amplified by PCR. These representations were labeled and applied to ROMA custom microarrays with 390,000 probes designed to detect DpnII fragments of appropriate size across the genome. The idea was to test how reproducible this two step process would be when

only 2-6 chromosomes are present in each single cell preparation (SKBR3 is roughly triploid).

The results are shown in Figure 5, presenting a comparison of the segmented profiles of a batch preparation of SKBR3 DNA (Figure 5A) against our normal male reference cell line (SKN-1) and three independent single cell DNAs (Figure 5 B,C,D). A visual inspection of these profiles makes it clear that after segmentation, the results for single cells are virtually identical to a standard DNA, leading us to have confidence that sequencing DNA from a representation will yield useful results. Although the raw ratio data is not presented here, we note that the standard deviation of the ratio data is much higher for the three single cell experiments (0.706, 0.725, 0.676) than for a batch DNA preparation (0.313). The extra noise does not adversely affect segmentation, although in certain regions very fine detail may be lost (red arrow in Figure 5A). This is comparable to other WGA1 experiments such as the one depicted in gray in Figure 4. In this light it is also worth noting that our particular use of the WGA1 method does not depend on absolutely uniform amplification across the genome. We benefit from the redundancy of data that we gain from segmentation to identify breakpoints. Even if only half of the potential fragments from a single cell were amplified in a given reaction, we would still obtain useful data.

Sequencing across chromosome breakpoints

One of the methods for the detection of rare tumor cells, in lymph nodes, blood or other bodily fluids is the amplification or sequencing of unique breakpoints resulting from deletions and/or translocations that occur during breast tumor development. Although events such as deletions and duplications are apparent from array CGH, the actual structure underlying the copy number changes is not.

The DNA sequences surrounding 22 breakpoints from two breast cancer cell lines were enriched by hybrid capture on custom microarrays as described by Hodges et al (20). Following hybrid capture, the enriched DNA was amplified and prepared for 'paired end' sequencing on the Illumina GA2 instrument. Paired end sequencing yields approximately 36 bp of sequence from each end of a single DNA molecule (after preparation for sequencing, each molecule is approximately 200 bp in length), and those sequences are then mapped back to the consensus genome. For a molecule from a normal, non-rearranged region the two matched reads should map approximately 130 bp apart. In contrast, at the edges of a deletion, the two reads from a single molecule would be separated on the map by the width of the deletion, or in the case of a translocation, would map to another chromosome. These are referred to as 'bridging fragments' because they connect regions not normally adjacent to one another. Examples of two such bridging fragments are shown at the top of Figure 6, along with the original CGH pattern, and graphs of the distance between paired ends (blue) and the total number of reads (red) across the capture region (X axis). By mapping the exact location of a breakpoint, highly sensitive and specific probes that will distinguish tumor cells from large numbers of normal cell can be prepared. Of 22 regions displaying deletions captured from cell lines T47D and MDA-MB-436, 15 behaved as simple deletions from which unique 'bridging fragments' could be identified as in Figure 6. Two additional sites 'jumped' to different chromosomes indicating translocations. The other 5 sites involved repeated sequences on one of the paired for which the identification of the exact breakpoint was not possible and which would not be suitable for creating unique probes.

Copy number profiling using varietal tags on restriction enzyme fragments.

As one illustration of varietal counting, copy number information was obtained using a "total different tagged nucleic acid molecules method.", DNA fragments were first associated with tags by using terminal transferase and nick-ligation (Figure 9). Genome DNA was obtained from the cell line SKBR3. DNA (2µg) was digested by NlaIII, resulting DNA fragments with 4bp, "CATG", 3' overhangs. This 3'OH end was extended with terminal transferase by adding a poly-A tail. One of primer adaptors was designed to have tags, 12 Ts and CATG at the 3' end, so it could be annealed to the 3' ends of the DNA fragments with a nick site (Figure 9, Product 3). This nick site was ligated by DNA ligase, forming duplex DNA fragments with tags. Another primer adaptor was then added to these duplex DNA fragments by one cycle of PCR (Figure 9, Step 5), allowing the resulting products of this reaction to be further amplified by PCR, followed by sequencing (Illumina/Solexa). Then, by mapping the subsequence of each tag associated sequence read to a location on the genome, the number of DNA fragments (assigned to the same genomic location) with different tags was counted.

We have previously obtained copy number profiles of SKBR3 genomic DNA from a million cell library, by sequencing and sequence counting. Figure 11 compares the use of varietal counting (a "total different tagged nucleic acid molecules method") with 'sequence counting' for analyzing copy number data, genome-wide (Figure 11A) and partial-genome (Figure 11B). The essentially equivalent result obtained using varietal counting illustrates that by adding varietal tags to DNA fragments before PCR one can use varietal counting as described herein to reliably obtain copy number information unaffected by amplification distortion.

Data analysis for the procedure described above employed a modification of the 'bin-counting' method because the NlaIII restriction sites are not distributed evenly in the genome. Therefore, in order to ensure equal weight for each of the 50K bins, the bin boundaries for this experiment were designed to encompass and equal number of NlaIII fragments sized between 100bp and 250bp, the size range from which the Illumina libraries were made.

Discussion

Single cell data is much more informative than data obtained from multiple cells, for example from array hybridization of multiple sections of a primary tumor. First, one can afford to analyze many more genomes by multiplex single cell sequencing than by array hybridization (on the order of a hundred single cells versus a dozen sections). Second, one is able to directly assess the exact proportion of cells in a region of a tumor that are malignant. Third, one is able to make a far more detailed analysis of the temporal order of genetic lesions, since one will not be looking at the average state of a population. Fourth, one is able to assess whether tumor heterogeneity is of the anatomically segregated or the co-habitation type, or both.

The invention described herein may be used to determine the original genome copy number (or the gene expression profile) of a sample without amplification distortion. All measurements of copy number or gene expression, especially involving multiple samples, are potential applications. Samples can be nucleic acids derived from single cells or populations of cells. The invention can be used to study cellular heterogeneity in tumors or normal populations of cells.

The invention may be used to generate a genomic profile of a cell, e.g., a cancer cell, or population of cells. From the

genomic profile it can be determined if specific genes are amplified or deleted. When the sample is derived from expressed RNA, and the regions are expressed genes, the relative counts comprise an expression profile. From the expression profile the tissue of origin of a cancer cell can be identified. When the sample is a mixture of genomes from different species, and the genomic regions distinguish the different species, the relative counts comprise a census of those different species. For example, from a census we can determine the population of microbial flora and relate the same to disease.

One of the advantages of the use of tags is that it allows the use of restriction endonuclease fragments in genome analysis. Counting methods for genome copy number determination use counts of independent reads. Two reads are not considered independent if they are the result of DNA amplification occurring during sample processing. Typically, this entails shearing sample DNA fragments, and the position of ends of the fragments are used to disambiguate PCR duplicates. If two DNA reads have the same ends, they could be PCR duplicates, and not independent, so they are counted as only one true read. One cannot disambiguate restriction fragments this way, because two reads of the same fragment will have the same ends. However, by the addition of tags, independent reads can be distinguished, if they are added before any amplification step, as they will have different tags. By contrast, if two reads of the same genome region have the same tag, they are highly likely to be PCR duplicates, and not independent.

The ability to use sequence of restriction fragments in genome analysis can be exploited to reduce the cost of determining copy number profile, much as performing ROMA facilitates genome analysis. By size selection of fragments, one can sample the genome at greater depth at fewer loci, and still obtain a high resolution copy number profile but with fewer

reads and therefore at lower cost. This is the essence of representation, and results in complexity reduction, the central element of ROMA. In ROMA, the analysis of the representation is done by hybridization to arrays, and here the representation is analyzed by sequencing.

Further complexity reduction can be achieved by sequence capture. The sequence capture can be designed specifically for the fragments that can be predicted by in silico methods to map to the regions where one wants copy number mutation. In this way one can target special regions of the genome, such as the exome, or known cancer genes. Very little sequence of the sample is then needed for copy number determination, and samples can be multiplexed at a great cost saving.

The invention disclosed herein may also be used to determine the proportion of components in a mixture. For example, suppose one possesses a set of reagents, such as antibody molecules, that bind to a set of substrates, such as different proteins, and one desires to detect the relative proportion of those substrates in a sample, such as blood. First, one covalently combines a tag having a sample tag with each reagent, where the sample tag portion identifies the reagent, and the varietal tag will be used in counting. The tagged reagents are then combined with the sample, and reagents that combine with substrates are physically separated. For example, the substrates may be biotinylated before being mixed with reagents, then reagents combining with substrates separated by avidin affinity chromatography. The tags are amplified by PCR and sequenced, effectively yielding relative counts for each reagent species and therefore each substrate. In this case, the constant portions of the tags include the sample identifier and nucleotides used for amplification.

The invention provides the following advantages: (1) the method is simple; (2) with statistical analysis the confidence

of a measurement can be determined with great accuracy; (3) this is a sequence based method, which in the long run will be cheaper than arrays; (4) long read lengths are not necessary, providing savings; (5) because tagged nucleic acid molecules are counted, many processing methods that create distortion in yields can be applied without major loss in the tagged nucleic acid molecule count; (6) among these processing methods are hybrid selection (aka capture microarrays), which restrict the sequence information to regions of importance, and thus reduce cost by reducing the total number of reads per sample needed for determination of the desired information; (7) the molecules from one sample can be combined with tags and a sample tag, creating sample tagged nucleic acid molecules which can then be pooled, and after processing and sequencing, the sample tags decoded, giving a profile for each sample; (8) pooling adds efficiency and uniformity; (9) the samples can be individual cells, yielding large numbers of single cell measurements cheaply.

The invention described herein has clinical applications as described above, including: (1) determination of the genomic heterogeneity of tumors; (2) detection of tumor cells in needle biopsies, in particular an assessment of the heterogeneity of the cells in a biopsy; (3) assessment of the spread of a cancer to regional lymph nodes; and (4) detection of malignant cells in sites such as blood and bone marrow. The presence of cells with the molecular signature of the primary in the blood or bone marrow opens up the possibility of using more general single cell/molecule assays as a first line of defense in the early detection of cancer, i.e. breast cancer in women at risk.

When there is radiological evidence of a breast mass, the patient most commonly undergoes fine needle biopsy, which is examined by the pathologist who makes an assessment of the

malignancy of the mass. Because needle aspiration destroys the histology of the biopsy, one of the pathologist's canonical tools for assessing malignancy is not available. Single cell sequencing would not only more accurately predict the presence of malignant cells, but also give what we might call "genomic staging." Is the tumor a simplex type of complex type? Is it mongenomic or polygenomic? Are there critical loci that are amplified or deleted that might immediately suggest a preoperative therapy?

The single cell/molecule approach allows the assessment of the spread of cancer to regional lymph nodes and to establish the DNA-methylation states at switch regions.

Successful application of single cell/molecule detection in the above contexts will lead to application to the detection of cancer recurrence, and the measurement of cancer load. Detection of recurrence at the earliest possible time may prolong survival for a large class of patients by earlier intervention than they would otherwise receive. Moreover, if cancer load were monitored in the patient, even before the site of the cancer growth is discernible by imaging, then one could measure the response of the cancer to a variety of therapeutic measurements. That is, the most effective choice of therapy could be determined empirically in the patient.

Additional applications exist in the diagnosis and selection of treatment for patients with auto-immune diseases, and in vitro fertilization (IVF) screening of pre-implant embryos for genetic defects such as trisomy-21.

References

The disclosures of the following references in their entireties are hereby incorporated by reference into this application.

1. Pinard R, de Winter A, Sarkis GJ, Gerstein MB, Tartaro KR, Plant RN, et al. Assessment of whole genome amplification-induced bias through high-throughput, massively parallel whole genome sequencing. BMC Genomics. 2006; 7:216. PMCID: PMC1560136.

2. Klein, C. et al. Comparative genomic hybridization, loss of heterozygosity, and DNA sequence analysis of single cells. PNAS. 1999; 96, pp. 4494-4499.

3. Stoecklein, N. et al. SCOMP Is Superior to Degenerated Oligonucleotide Primed-Polymerase Chain Reaction for Global Amplification of Minute Amounts of DNA from Microdissected Archival Tissue Samples. American Journal of Pathology. 2002; 161(1):43-51.

4. Parameswaran et al. A pyrosequencing-tailored nucleotide barcode design unveils opportunities for large-scale sample multiplexing. Nucleic Acids Research. 2007; 35(19): e130.

5. U.S. Patent No. 7,622,281, Ronaghi et al.

6. Eid et al. Real-Time DNA Sequencing from Single Polymerase Molecules. Science. 2009; 323:133-138.

7. U.S. Patent No. 5,639,603, Dower et al.

8. U.S. Patent Application Publication No. 2006/0073506, Christians et al.

9. Navin N, Krasnitz A, Rodgers L, Cook K, Meth J, Kendall J, et al. Inferring tumor progression from genomic

heterogeneity. Genome Res. 2010;20(1):68-80. PMCID: 2798832.

10. Fisher B, Redmond CK, Fisher ER. Evolution of knowledge related to breast cancer heterogeneity: a 25-year retrospective. J Clin Oncol. 2008;26(13):2068-71.

11. Langmead B, Trapnell C, Pop M, Salzberg SL. Ultrafast and memory-efficient alignment of short DNA sequences to the human genome. Genome Biol. 2009;10(3):R25. PMCID: PMC2690996.

12. Saitou N, Nei M. The neighbor-joining method: a new method for reconstructing phylogenetic trees. Mol Biol Evol. 1987;4(4):406-25.

13. De Baetselier P, Roos E, Brys L, Remels L, Gobert M, Dekegel D, et al. Nonmetastatic tumor cells acquire metastatic properties following somatic hybridization with normal cells. Cancer Metastasis Rev. 1984;3(1):5-24.

14. Duelli DM, Padilla-Nash HM, Berman D, Murphy KM, Ried T, Lazebnik Y. A virus causes cancer by inducing massive chromosomal instability through cell fusion. Curr Biol. 2007;17(5):431-7.

15. Jorgensen HF, Adie K, Chaubert P, Bird AP. Engineering a high-affinity methyl-CpG-binding protein. Nucleic Acids Res. 2006;34(13):e96. PMCID: PMC1540740.

16. Meehan RR, Lewis JD, Bird AP. Characterization of MeCP2, a vertebrate DNA binding protein with affinity for methylated DNA. Nucleic Acids Res. 1992;20(19):5085-92. PMCID: PMC334288.

17. Huang J., Pang J., Watanabe T., Ng HK, Ohgaki H. Whole genome amplification for array comparative genomic hybridization using DNA extracted from formalin-fixed,

paraffin-embedded histological sections. J Mol Diagn. 2009 Mar;11(2):109-16. Epub 2009 Feb 5.

18. Talseth-Palmer BA, Bowden NA, Hill A, Meldrum C, Scott RJ. Whole genome amplification and its impact on CGH array profiles. BMC Res Notes. 2008 Jul 29;1:56.

19. Hodges E, Smith AD, Kendall J, Xuan Z, Ravi K, Rooks M, Zhang MQ, Ye K, Bhattacharjee A, Brizuela L, McCombie WR, Wigler M, Hannon GJ, Hicks JB. High definition profiling of mammalian DNA methylation by array capture and single molecule bisulfite sequencing. Genome Res. 2009 Sep;19(9):1593-605. Epub 2009 Jul 6.

20. Hodges E, Xuan Z, Balija V, Kramer M, Molla MN, Smith SW, Middle CM, Rodesch MJ, Albert TJ, Hannon GJ, McCombie WR. Genome-wide in situ exon capture for selective resequencing. Nat Genet. 2007 Dec;39(12):1522-7. Epub 2007 Nov 4.

21. genome.ucsc.edu/index.html?org=Human&db=hg19&hgsid=171216665

22. Miner B.E., Stoger, R.J., Burden, A.F., Laird, C.D., Hansen R.S.. Molecular barcodes detect redundancy and contamination in hairpin-bisulfite PCR. Nucleic Acids Research, 2004:32(17):e135.

23. U.S. Patent No. 7,537,897, issued May 26, 2009 (Brenner et al.).

24. McCloskey M.L., Stoger, R., Hansen, R.S., Laird, C.D. Encoding PCR Products with Batch-stamps and Barcodes. Biochem. Genet. 2007:45:761-767.

What is claimed is:

1. A method for obtaining from genomic material genomic copy number information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) tagging the segments with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises one segment of the genomic material from step (a) and a tag;

c) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

d) generating tag associated sequence reads by sequencing the product of step (c);

e) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome; and

f) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining genomic copy number information unaffected by amplification distortion.

2. The method of claim 1, further comprising estimating a genomic copy number of a region of the genome comprising more than one location on the genome by assigning as the copy number of the region the highest count obtained in step (f) for the locations within the region.

3. The method of claim 1, further comprising comparing a count obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

4. The method of claim 1, further comprising

a) summing the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

b) summing the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region is comprised of a number of locations which is comparable to the number of locations of the first region;

c) comparing the value obtained in step (a) to the value obtained in step (b),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

5. The method of claim 4, wherein step (b) further comprises

i) summing the counts obtained in step (f) for locations on the genome which comprise

a third region of the genome, wherein the third region is comprised of a number of locations which is comparable to the number of locations of the first region; and

ii) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (f) for locations which comprise the third region.

6. The method of any one of claims 4-5, wherein the second region of the genome comprises a centromere.

7. The method of claim 1, further comprising summing the counts obtained in step (f) for locations which comprise a region of the genome, and comparing the sum to a sum obtained from a reference sample for the same region of the genome, thereby estimating a relative genomic copy number of the region of the genome.

8. A method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA;

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises

i) one cDNA from step (a) or one segment of the cDNA from step (b), and

ii) a tag;

d) subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR);

e) generating tag associated sequence reads by sequencing the product of step (d);

f) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a cDNA or a segment of cDNA to a location on the cDNA library; and

g) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

9. A method for obtaining from mRNA transcripts mRNA copy number information unaffected by amplification distortion, comprising:

a) generating tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under

conditions that promote the formation of only one complement, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to the first order derivative strands; and

iii) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise substantially unique primers, thereby generating tagged nucleic acid molecules;

b) subjecting the tagged nucleic acid molecules to polymerase chain reaction (PCR) amplification;

c) generating tag associated sequence reads by sequencing the product of step (b);

d) assigning each tagged nucleic acid molecule to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read corresponding to a mRNA transcript to a location on the cDNA library; and

e) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information unaffected by amplification distortion.

10. A method for obtaining from genomic material DNA methylation information unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands to a polymerase reaction in the presence essentially unique primers, wherein the essentially unique primers are capable of hybridizing to the polynucleotide tail of the zero-th order strands, under conditions that promote the formation of only one complement, thereby generating tagged nucleic acid molecules;

d) separating the tagged nucleic acid molecules into a group consisting of hemi-methylated tagged nucleic acid molecules and a group consisting of unmethylated tagged nucleic acid molecules;

e) subjecting each group of step (d) to amplification by polymerase chain reaction (PCR);

f) generating tag associated sequence reads by sequencing the product of step (e);

g) assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome; and

h) counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

thereby obtaining DNA methylation information unaffected by amplification distortion.

11. The method of any one of claims 1-7, wherein tagging the segments to generate tagged nucleic acid molecules comprises:

a) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

b) subjecting the zero-th order derivative strands of step (a) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

c) adding a polynucleotide tail to the first order derivative strands;

d) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the

polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (b) and (d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

12. The method of any one of claims 1-7, wherein tagging the segments to generate tagged nucleic acid molecules comprises:

a) adding a polynucleotide tail to the ends of the segments of genomic material to generate zero-th order derivative strands,

b) subjecting the zero-th order derivative strands of step (a) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (a) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

c) subjecting the product of step (b) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (a) under conditions which promote the formation of only one complement, wherein the primers of step (c) have different nucleotide sequences than the primers of step (b), and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (b) and (c) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

13. The method of any one of claims 9-12, wherein adding a polynucleotide tail comprises the use of a terminal transferase.

14. The method of any one of claims 1-7, wherein tagging the segments to generate tagged nucleic acid molecules comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic material.

15. The method of claim 14, wherein the adaptors comprising the tags are ligated to only one end of the segments of the genomic material.

16. The method of any one of claims 1-15, wherein the tags comprise a sequence that aids PCR amplification.

17. The method of any one of claims 1-16, wherein each tagged nucleic acid molecule comprises one tag.

18. The method of any one of claims 1-17, wherein each tagged nucleic acid molecule comprises more than one tag.

19. The method of any one of claims 1-7 or 10-18, wherein segments of the genomic material are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

20. The method of claim 8, wherein segments of the cDNA are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

21. The method of any one of claims 8-9, 13, 17-18 or 20, wherein the maximum copy number of a location on a cDNA library is not less than the number of tagged nucleic

acid molecules having a different tag that have been assigned to the same location on the cDNA library.

22. The method of any one of claims 1-7 or 10-19, further comprising analyzing mRNA copy number.

23. The method of any one of claims 10, 13, 16-19, or 22, wherein separating the tagged nucleic acid molecules into a group consisting of hemi-methylated tagged nucleic acid molecules and a group consisting of unmethylated tagged nucleic acid molecules is by cleavage with methylation sensitive restriction enzymes, partitioning with antibodies, or partitioning with methyl-C binding proteins directed to methylated or hydroxymethylated cytosine.

24. The method of any one of claims 1-23, wherein the tagged nucleic acid molecules are subject to hybrid capture prior to PCR or prior to sequencing.

25. The method of any one of claims 1-24, wherein each tagged nucleic acid molecule differs at more than one nucleotide.

26. The method of any one of claims 1-25, wherein the tag sequences further comprise a sample tag.

27. The method of claim 26, wherein the tagged nucleic acid molecules are pooled with a plurality of tagged nucleic acid molecules having a different sample tag prior to PCR amplification or prior to sequencing.

28. The method of claim 27, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads according to sample tag.

29. The method of any one of claims 1-28, wherein the tagged nucleic acid molecules are generated from a single species.

30. The method of claim 29, wherein the tagged nucleic acid molecules are generated from a single organism.

31. The method of any one of claims 1-30, wherein tagged nucleic acid molecules are generated from a single cell.

32. The method of claim 31, wherein the single cell is from a needle aspirate of suspected cancer lesions.

33. The method of claim 31, wherein the single cell is from a core biopsy of suspected cancer lesions.

34. The method of claim 29, wherein the tagged nucleic acid molecules are generated from two or more organisms.

35. The method of claim 34, wherein the tagged nucleic acid molecules are generated from a single cell of each organism.

36. The method of any one of claims 30-35, wherein the single species is human.

37. The method of any one of claims 1-28, wherein the tagged nucleic acid molecules are generated from two or more species.

38. The method of claim 37, wherein the tagged nucleic acid molecules are generated from a population of microbes.

39. The method of claim 38, comprising comparing the genomic copy number information obtained for different species of the population to determine the relative count of those different species in the population.

40. A composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of the genomic material;

b) adding a polynucleotide tail to the ends of the segments of the genomic material to generate zero-th order derivative strands;

c) subjecting the zero-th order derivative strands of step (b) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

d) adding a polynucleotide tail to the first order derivative strands;

e) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (c) and (e) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

41. A composition of matter derived from genomic material comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining segments of genomic material;

b) adding a polynucleotide tail to the ends of the segments of genomic material to generate zero-th order derivative strands,

c) subjecting the zero-th order derivative strands of step (b) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (b) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

d) subjecting the product of step (c) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (b) under conditions which promote the formation of only one complement, wherein the primers of step (d) have different nucleotide sequences than the primers of step (c), and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (c) and (d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

42. A composition of matter derived from mRNA transcripts comprising tagged nucleic acid molecules, said tagged

nucleic acid molecules being produced by a process comprising:

a) obtaining cDNA from the mRNA transcripts;

b) optionally obtaining segments of the cDNA; and

c) tagging the cDNA or segments of cDNA with substantially unique tags to generate tagged nucleic acid molecules, wherein each tagged nucleic acid molecule comprises

iii) one cDNA from step (a) or one segment of the cDNA from step (b) and

iv) a tag.

43. A composition of matter derived from mRNA transcripts comprising tagged nucleic acid molecules, said tagged nucleic acid molecules being produced by a process comprising:

a) obtaining mRNA transcripts;

b) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement, thereby generating first order derivative strands;

c) adding a polynucleotide tail to the first order derivative strands;

d) subjecting the first order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the

polynucleotide tail added in step (c) under conditions that promote the formation of only one complement, thereby generating second order derivative strands,

wherein the primers of at least one of steps (b) and d) comprise substantially unique primers, thereby generating tagged nucleic acid molecules.

44. The composition of matter of any one of claims 40-41 or 43, wherein adding a polynucleotide tail comprises the use of a terminal transferase.

45. The composition of matter of any one of claims 40-44, wherein each tagged nucleic acid molecule comprises one tag.

46. The composition of matter of any one of claims 40-44, wherein each tagged nucleic acid molecule comprises more than one tag.

47. The composition of matter of any one of claims 40-41 or 44-46, wherein segments of the genomic material are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

48. The composition of matter of any one of claims 42-46, wherein segments of the cDNA are produced by restriction endonuclease digestion, mechanical shearing, heating, or sonication.

49. The composition of matter of any one of claims 40-48, wherein the complexity of the tagged nucleic acid molecules is reduced by hybrid capture.

50. The composition of matter of any one of claims 40-49, wherein each tagged nucleic acid molecules differs at more than one nucleotide.

51. The composition of matter of any one of claims 40-50, wherein the tag sequences further comprise a sample tag.

52. A composition of matter comprising at least two pools of tagged nucleic acid molecules according to claim 51.

53. The composition of matter of any one of claims 40-52, wherein the tagged nucleic acid molecules are generated from a single cell.

54. The composition of matter of claim 53, wherein the single cell is from a needle aspirate of suspected cancer lesions.

55. The composition of matter of claim 53, wherein the single cell is from a core biopsy of suspected cancer legions.

56. The composition of matter of any one of claims 40-52, wherein the tagged nucleic acid molecules are generated from two or more organisms.

57. A kit for determining nucleic acid copy number information unaffected by amplification distortion comprising:

a) a terminal transferase, and

b) plurality of substantially unique primers,

wherein the substantially unique primers comprise substantially unique tags, and wherein the substantially unique primers are capable of hybridizing to the polynucleotide tail of a nucleic acid molecule which has a polynucleotide tail added by the terminal transferase.

58. The kit of claim 57, further comprising a DNA polymerase having 3′-5′ proofreading activity.

59. The kit of any one of claims 57-58, wherein the plurality of substantially unique primers comprises 10^n primers, wherein n is an integer from 2 to 9.

60. The kit of any one of claims 57-59, further comprising a second terminal transferase and a primer capable of hybridizing to the polynucleotide tail of a nucleic acid molecule which has a polynucleotide tail added by the second terminal transferase.

61. The kit of any one of claims 57-60, wherein the tags are six nucleotides long.

62. The kit of any one of claims 57-60, wherein the tags are 15 nucleotides long.

63. The kit of any one of claims 57-62, further comprising sample tags.

64. The kit of claim 63, wherein the sample tags are 2 nucleotides long.

65. The kit of claim 63, wherein the sample tags are 4 nucleotides long.

66. The kit of any one of claims 63-65, wherein the sample tags comprise a sample tag set.

67. The kit of any one of claims 63-66, wherein the substantially unique tags comprise the sample tags.

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Abstract of the Disclosure

A method for obtaining from genomic material genomic copy number information unaffected by amplification distortion, comprising obtaining segments of the genomic material, tagging the segments with substantially unique tags to generate tagged nucleic acid molecules, such that each tagged nucleic acid molecule comprises one segment of the genomic material and a tag, subjecting the tagged nucleic acid molecules to amplification by polymerase chain reaction (PCR), generating tag associated sequence reads by sequencing the product of the PCR reaction, assigning each tagged nucleic acid molecule to a location on a genome associated with the genomic material by mapping the subsequence of each tag associated sequence read corresponding to a segment of the genomic material to a location on the genome, and counting the number of tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome, thereby obtaining genomic copy number information unaffected by amplification distortion.

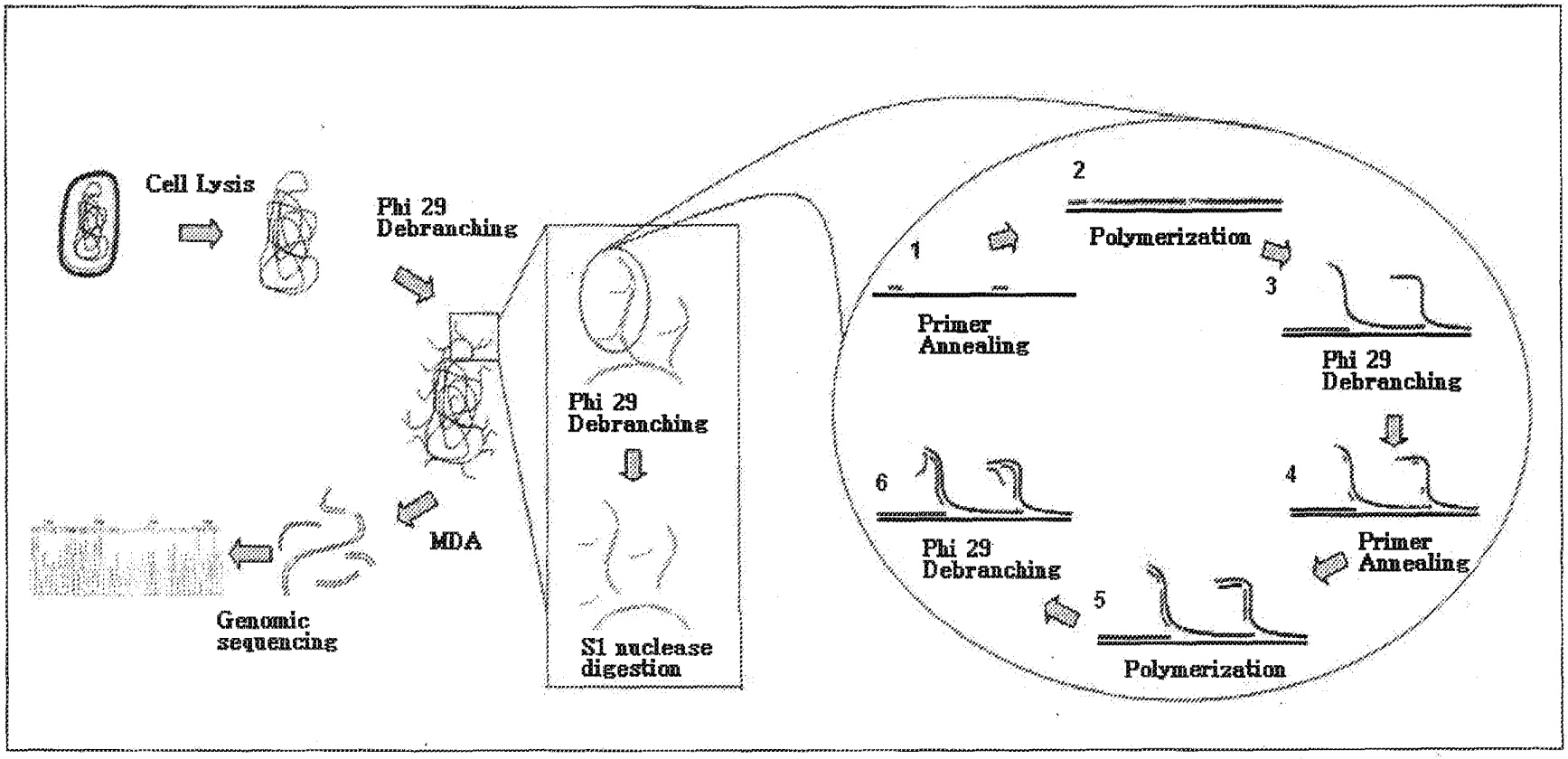

Cell Lysis
Phi 29
Debranching
MDA
Genomic
sequencing
Phi 29
Debranching
S1 nuclease
digestion
1
Primer
Annealing
2
Polymerization
3
Phi 29
Debranching
4
Primer
Annealing
5
Polymerization
6
Phi 29
Debranching

Fig. 1

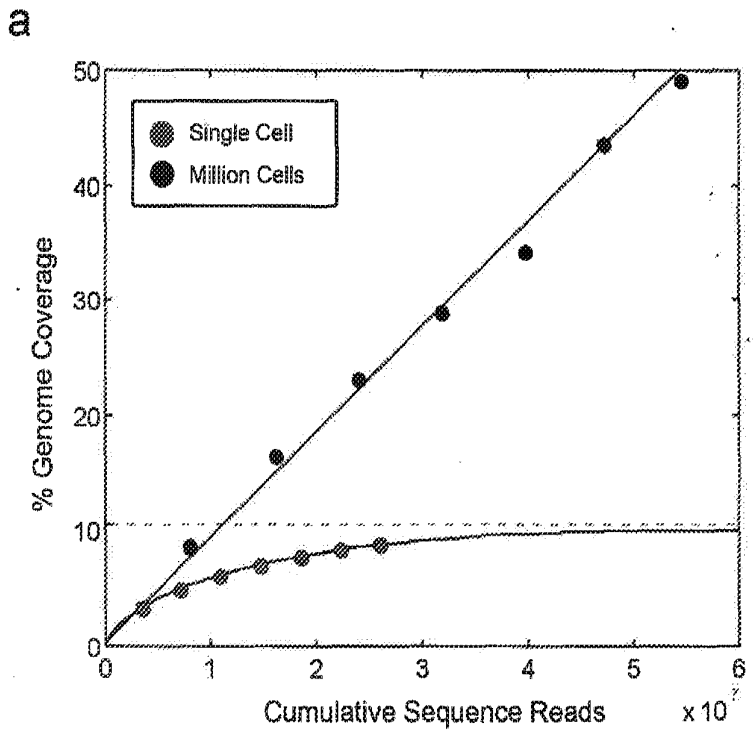

a
Single Cell
Million Cells
% Genome Coverage
Cumulative Sequence Reads
x 10^7
0
10
20
30
40
50
0
1
2
3
4
5
6

Fig. 2

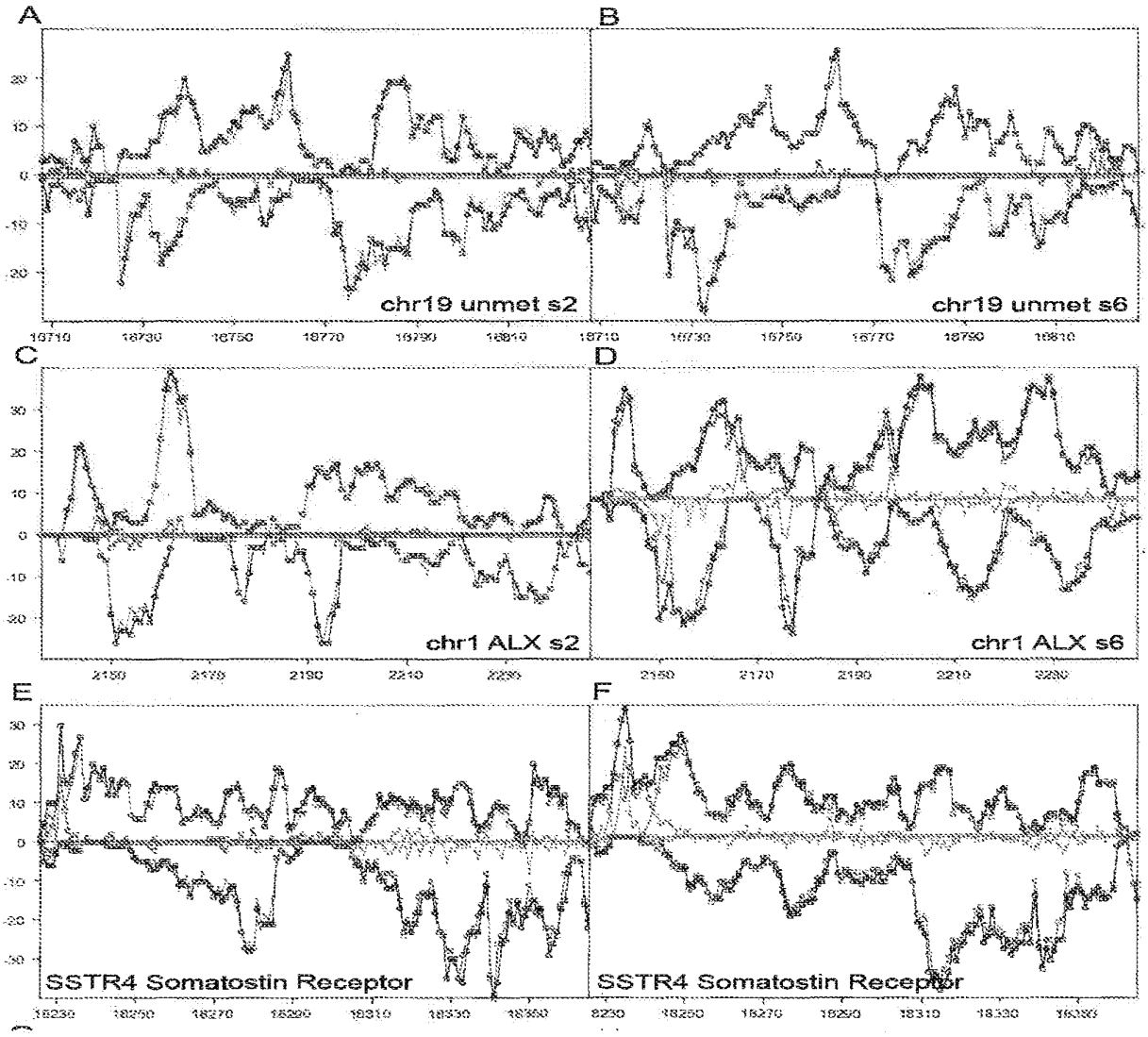

A
chr19 unmet s2
B
chr19 unmet s6
C
chr1 ALX s2
D
chr1 ALX s6
E
SSTR4 Somatostin Receptor
F
SSTR4 Somatostin Receptor

Fig. 3

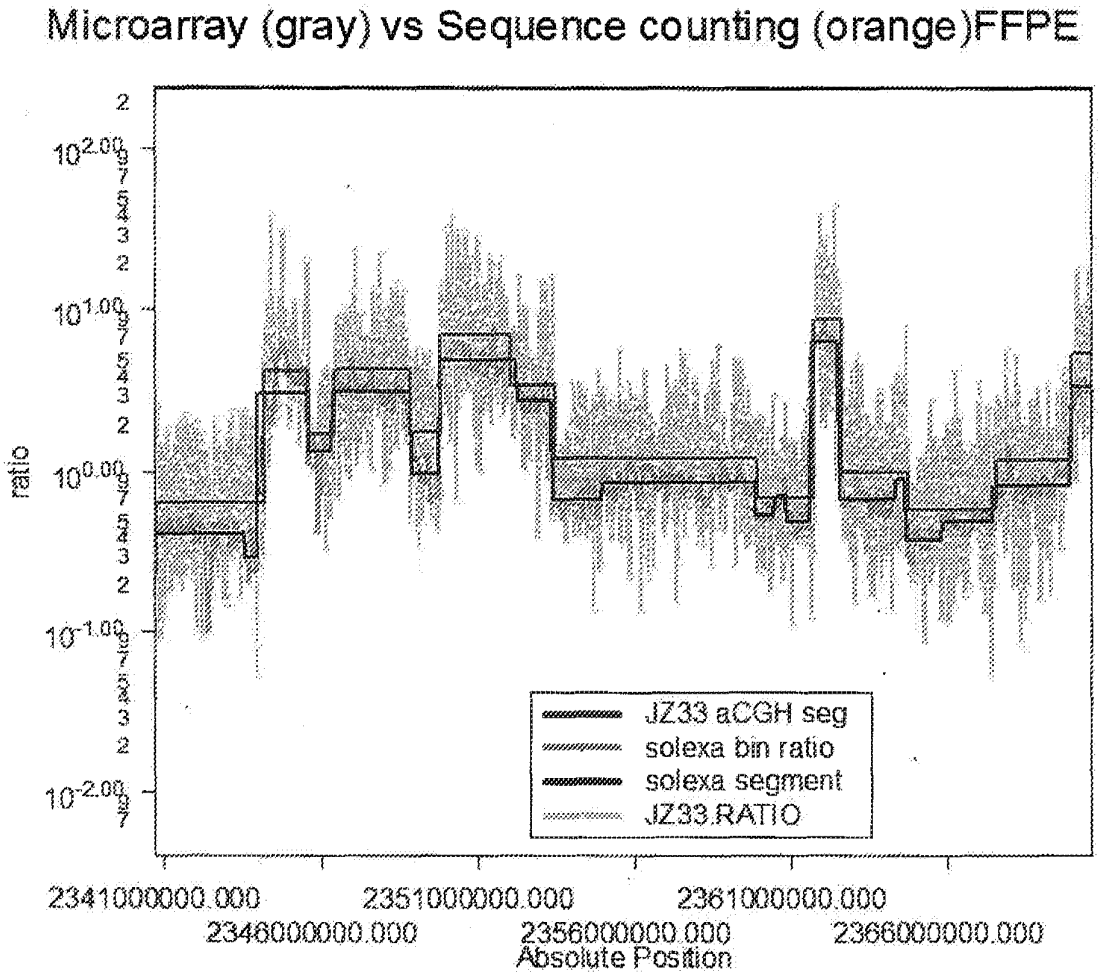

Microarray (gray) vs Sequence counting (orange)FFPE JZ33
ratio
JZ33 aCGH seg
solexa bin ratio
solexa segment
JZ33.RATIO
2341000000.000
2346000000.000
2351000000.000
2356000000.000
2361000000.000
2366000000.000
Absolute Position

Fig. 4

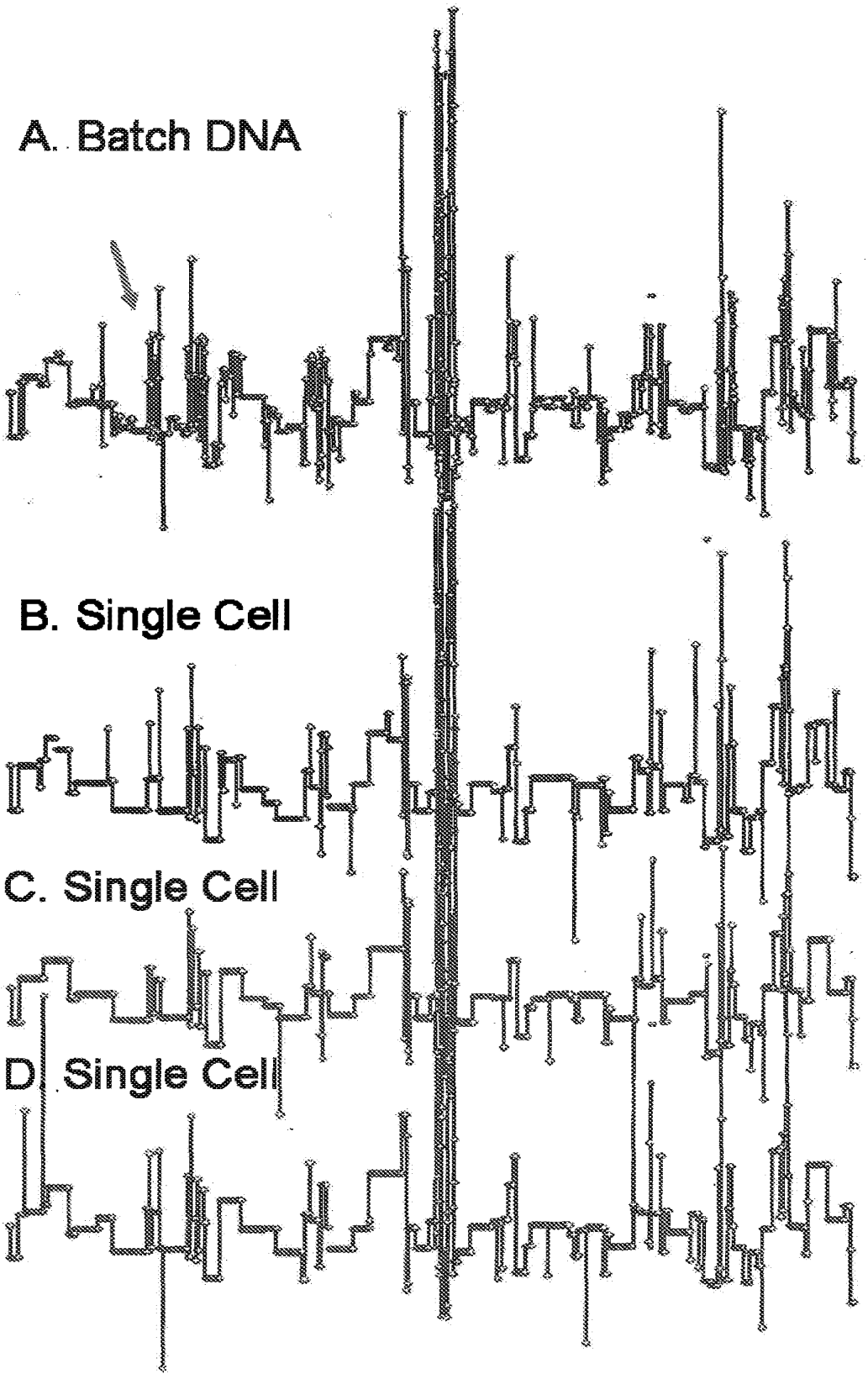


Fig. 5

GUARDANT HEALTH EX.1009.116

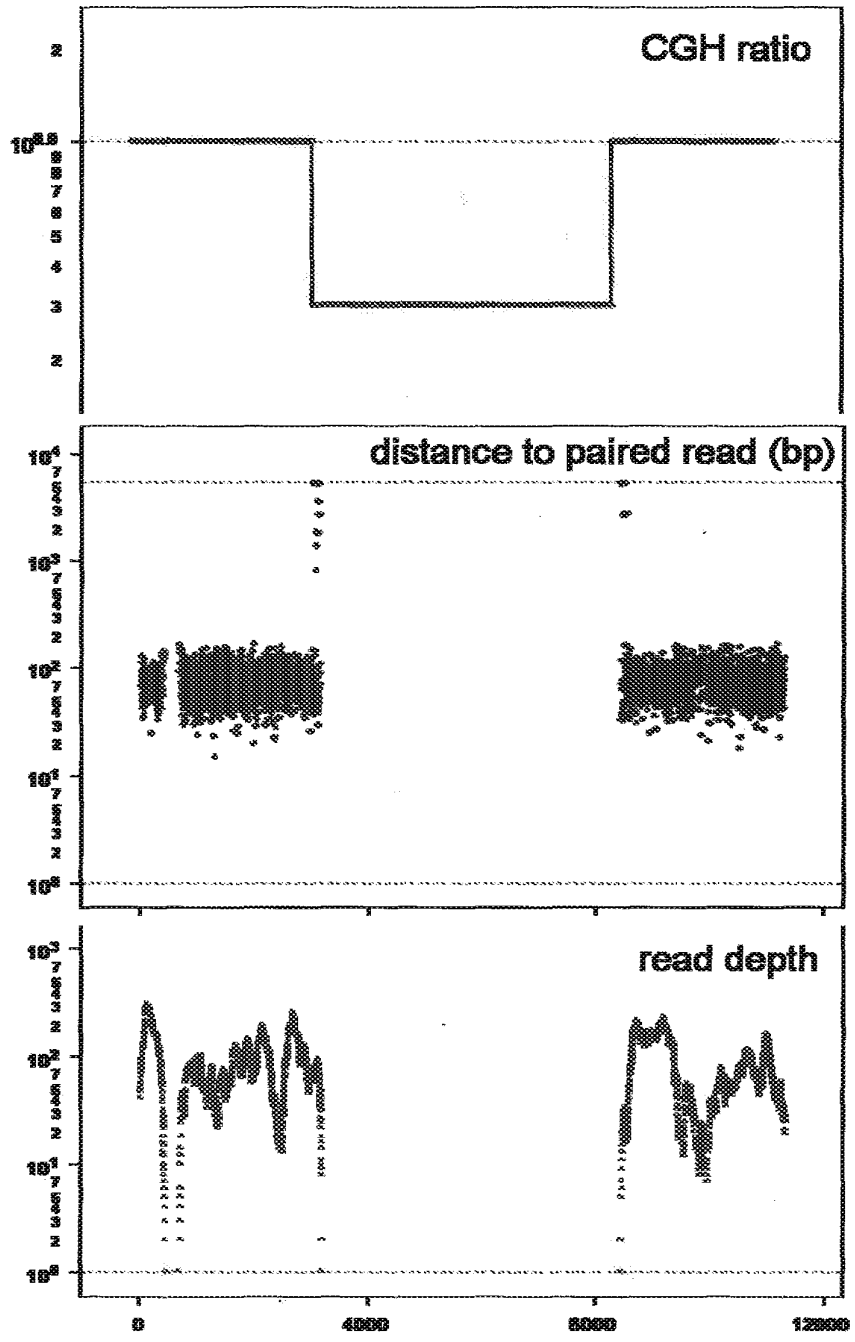

CGH ratio
distance to paired read (bp)
read depth
0
4000
8000
12000

Fig. 6

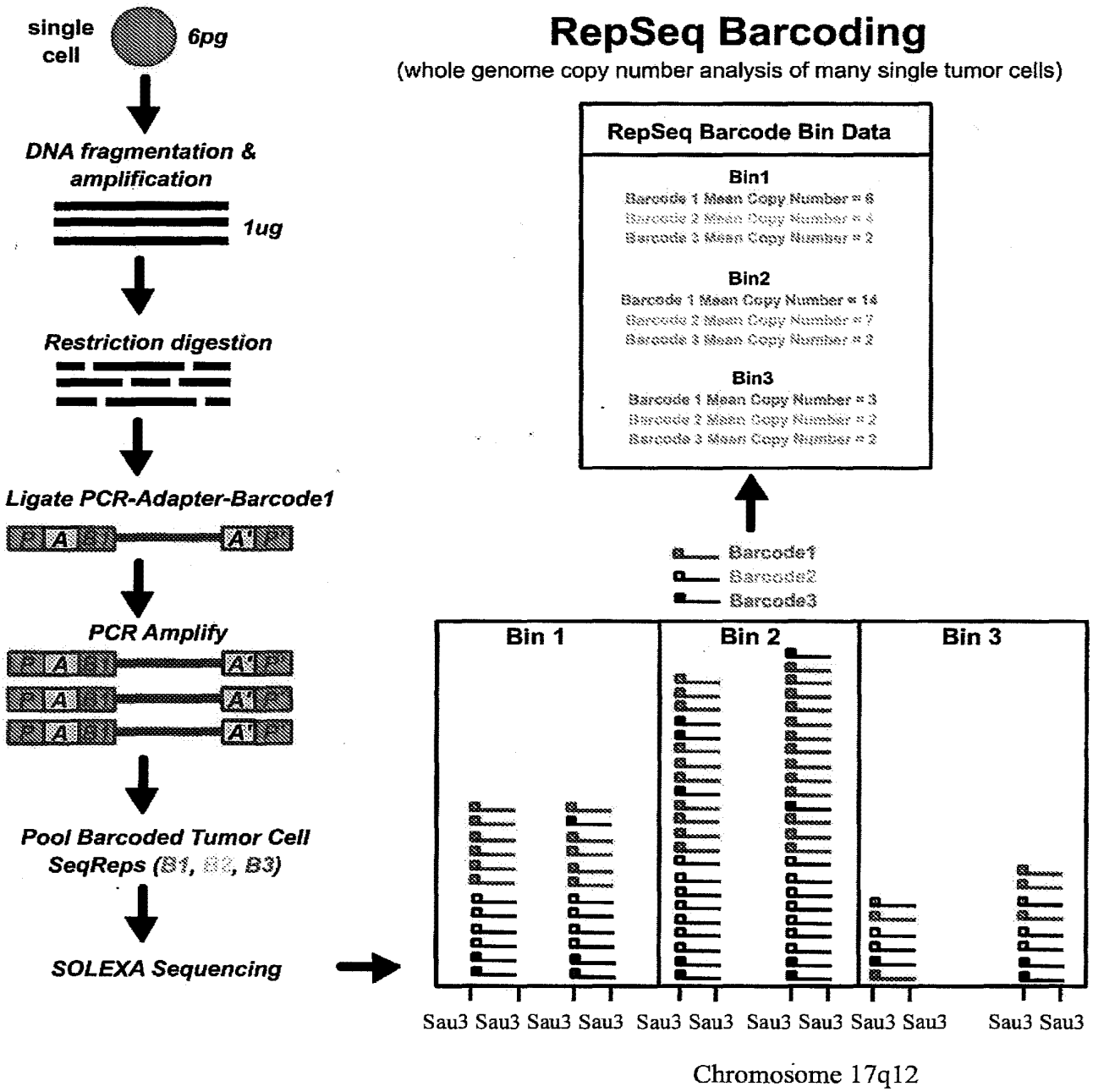

single cell
6pg
DNA fragmentation & amplification
1ug
Restriction digestion
Ligate PCR-Adapter-Barcode1
PCR Amplify
Pool Barcoded Tumor Cell SeqReps (B1, B2, B3)
SOLEXA Sequencing
RepSeq Barcoding
(whole genome copy number analysis of many single tumor cells)
RepSeq Barcode Bin Data
Bin1
Barcode 1 Mean Copy Number = 6
Barcode 2 Mean Copy Number = 4
Barcode 3 Mean Copy Number = 2
Bin2
Barcode 1 Mean Copy Number = 14
Barcode 2 Mean Copy Number = 7
Barcode 3 Mean Copy Number = 2
Bin3
Barcode 1 Mean Copy Number = 3
Barcode 2 Mean Copy Number = 2
Barcode 3 Mean Copy Number = 2
Barcode1
Barcode2
Barcode3
Bin 1
Bin 2
Bin 3
Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3 Sau3
Chromosome 17q12

Fig. 7

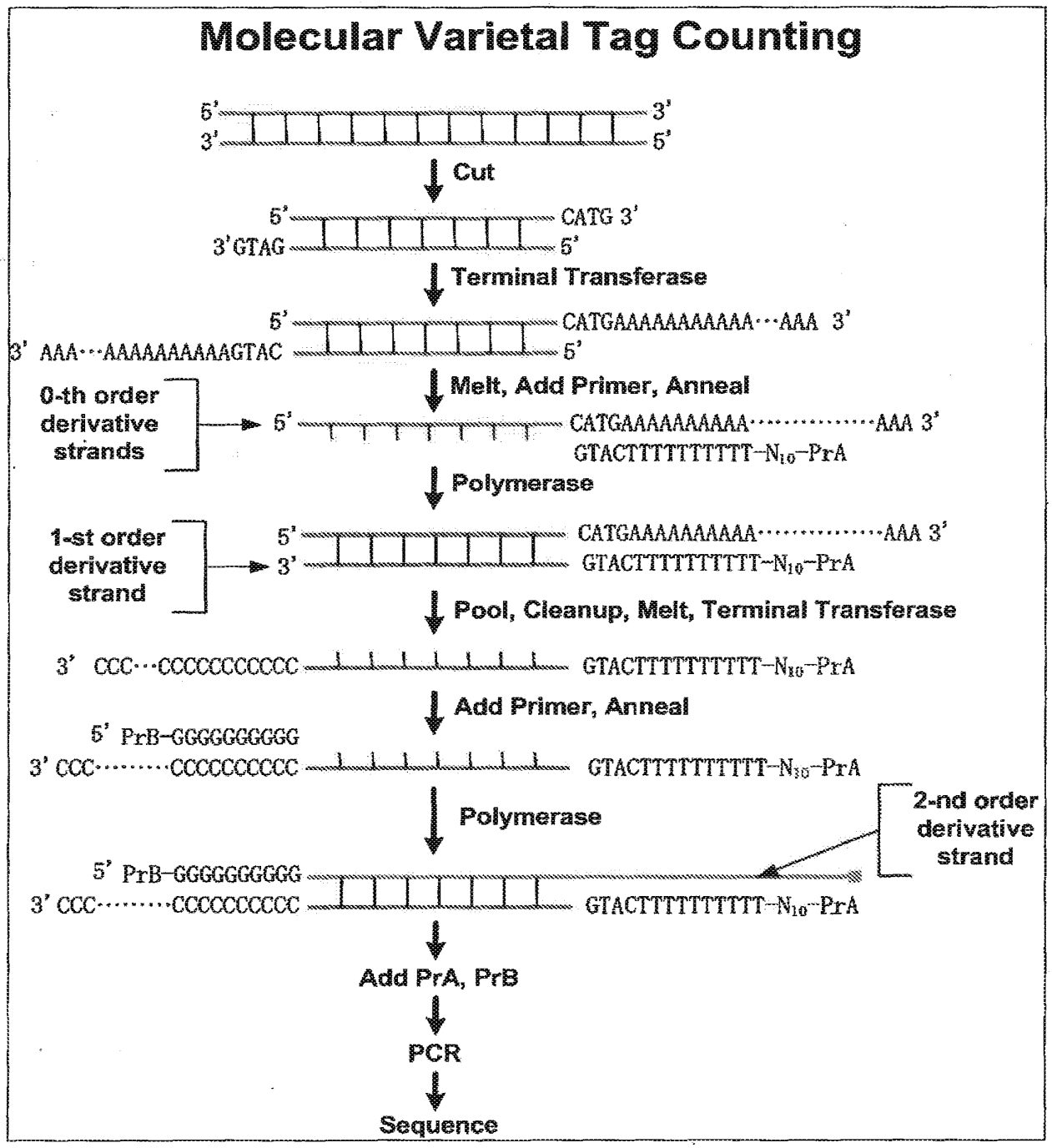

Molecular Varietal Tag Counting
5'
3'
3'
5'
Cut
5'
CATG 3'
3'GTAG
5'
Terminal Transferase
5'
CATGAAAAAAAAAAA···AAA 3'
3' AAA···AAAAAAAAAAGTAC
5'
Melt, Add Primer, Anneal
0-th order derivative strands
5'
CATGAAAAAAAAAA·····AAA 3'
GTACTTTTTTTTTT-N10-PrA
Polymerase
1-st order derivative strand
5'
3'
CATGAAAAAAAAAA·····AAA 3'
GTACTTTTTTTTTT-N10-PrA
Pool, Cleanup, Melt, Terminal Transferase
3' CCC···CCCCCCCCCC
GTACTTTTTTTTTT-N10-PrA
Add Primer, Anneal
5' PrB-GGGGGGGGGG
3' CCC·····CCCCCCCCCC
GTACTTTTTTTTTT-N10-PrA
Polymerase
2-nd order derivative strand
5' PrB-GGGGGGGGGG
3' CCC·····CCCCCCCCCC
GTACTTTTTTTTTT-N10-PrA
Add PrA, PrB
PCR
Sequence

Fig. 8

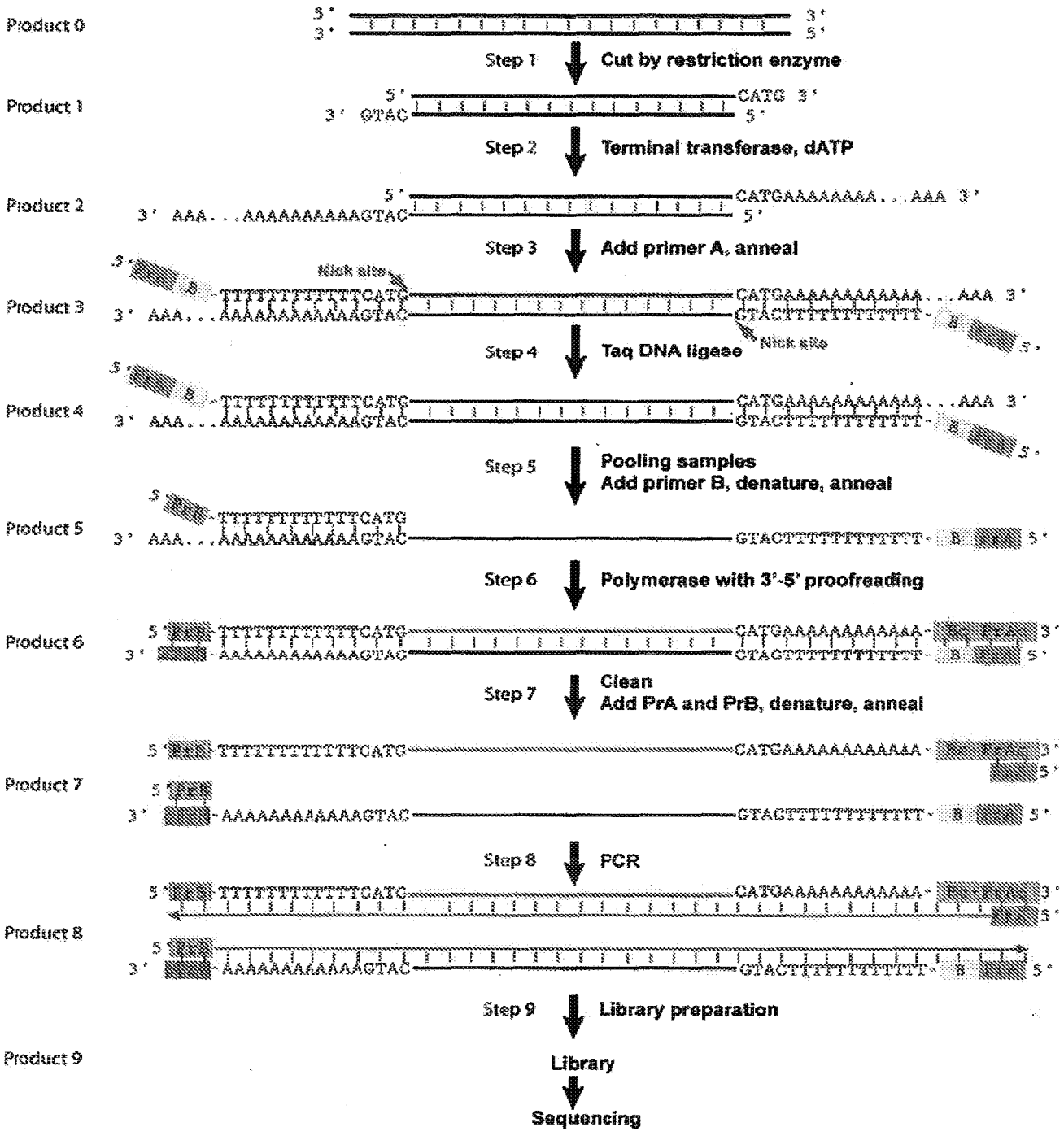

Product 0
Step 1
Cut by restriction enzyme
Product 1
Step 2
Terminal transferase, dATP
Product 2
Step 3
Add primer A, anneal
Nick site
Product 3
Step 4
Taq DNA ligase
Nick site
Product 4
Step 5
Pooling samples
Add primer B, denature, anneal
Product 5
Step 6
Polymerase with 3'-5' proofreading
Product 6
Step 7
Clean
Add PrA and PrB, denature, anneal
Product 7
Step 8
PCR
Product 8
Step 9
Library preparation
Product 9
Library
Sequencing

Fig. 9

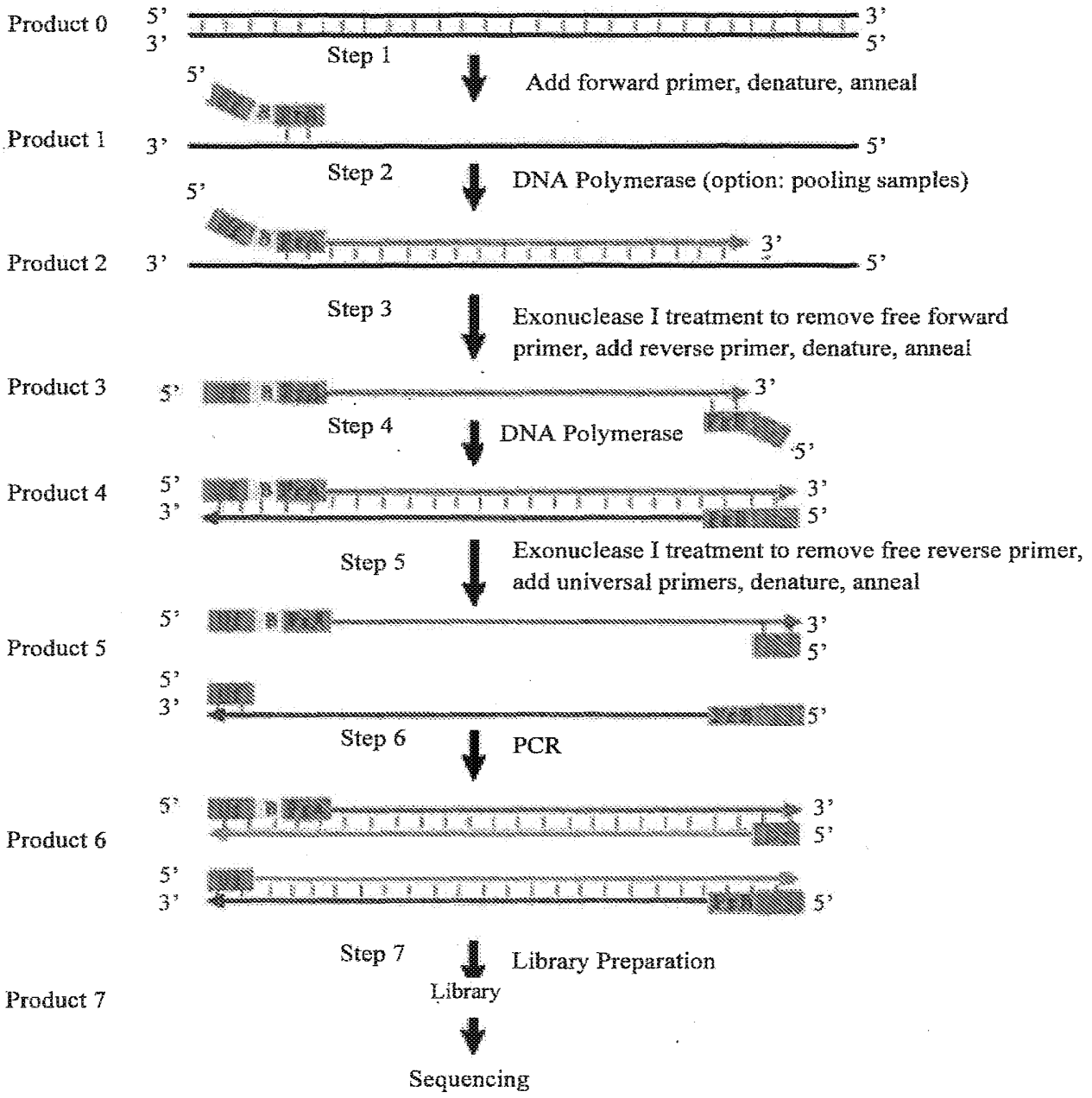

Product 0
5'
3'
3'
5'
Step 1
Add forward primer, denature, anneal
Product 1
Step 2
DNA Polymerase (option: pooling samples)
Product 2
Step 3
Exonuclease I treatment to remove free forward primer, add reverse primer, denature, anneal
Product 3
Step 4
DNA Polymerase
Product 4
Step 5
Exonuclease I treatment to remove free reverse primer, add universal primers, denature, anneal
Product 5
Step 6
PCR
Product 6
Step 7
Library Preparation
Library
Product 7
Sequencing

Fig. 10

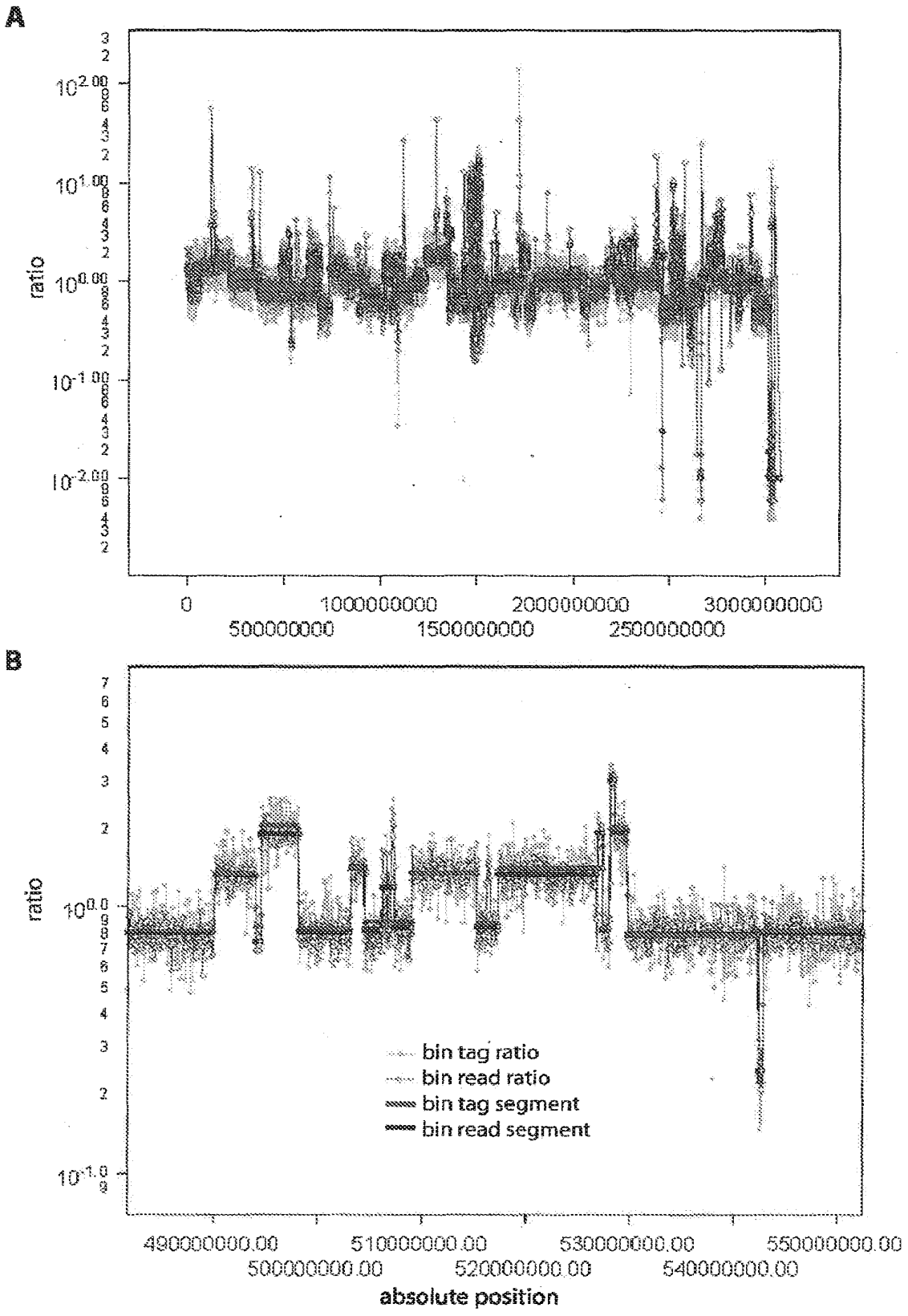

A
ratio
0
500000000
1000000000
1500000000
2000000000
2500000000
3000000000
B
ratio
bin tag ratio
bin read ratio
bin tag segment
bin read segment
490000000.00
500000000.00
510000000.00
520000000.00
530000000.00
540000000.00
550000000.00
absolute position

Fig. 11

Docket No. 81503-AA/GJG/AWG/AC

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.
U.S. Serial No.	:	Not Yet Known (continuation of U.S Serial No. 13/278,333, filed October 21, 2011)
Filed	:	Herewith
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza, 20th Floor
New York, New York 10112
March 7, 2016

BY EFS-WEB
Commissioner for Patents
Alexandria, VA

PETITION UNDER 37 CFR § 1.84(a)(2) TO ACCEPT COLORED FIGURES FILED IN CONNECTION WITH THE ABOVE-IDENTIFIED APPLICATION

This Petition is submitted under 37 CFR § 1.84(a)(2) to accept the colored Figures which are being filed herewith.

Fee for Filing a Petition under 37 CFR § 1.84(a)(2)

Pursuant to 37 CFR § 1.84(a)(2)(i), a fee of $70.00 is required for filing this Petition requesting acceptance of colored figures. Accordingly, authorization is hereby given to charge the amount of SEVENTY DOLLARS ($70.00) to Deposit Account No. 03-3125.

Color Drawings

The subject application is being filed with one (1) set of color drawings consisting of eleven (11) sheets containing Figures 1-11. Such color drawings are necessary because certain features of the Figures are referenced by color in the specification and/or would not be identifiable or distinguishable in black and white.

For example, Figures 3, 4, 5, and 11 show data in a graphical form which cannot be adequately represented in black and white.

Another example is Figures 8-10 which use colors to differentiate strands of nucleic acid molecules and to identify their source.

In accordance with the guidance set forth in Section H of "Legal Framework for Electronic Filing System-Web (EFS-Web)" [Federal Register 74:206 (October 27, 2009) pp. 55200-55211], the requirement for three (3) sets of color drawings or figures under 37 CFR § 1.84(a)(2)(ii) is not applicable to color drawings or figures submitted via EFS-Web. Therefore, applicants are submitting via EFS-Web only one (1) set of figures.

Reference to Color Figures in Specification

In accordance with 37 CFR §1.84(a)(2)(iii), the subject specification contains a paragraph immediately after the section heading titled "Brief Description Of The Figures" stating:

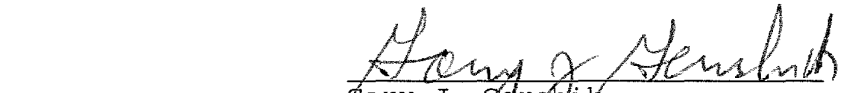
> "The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawing(s) will be provided by the Office upon request and payment of the necessary fee."

Applicants maintain that this Petition complies with the requirements of 37 CFR § 1.84(a)(2), and respectfully request that this Petition be granted and the sheets of colored figures filed with the subject application be accepted.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the enclosed filing fee of $730.00 and the abovementioned $70.00 fee for filing this Petition, is deemed necessary in connection with the filing of this Petition and the application it accompanies. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Docket Number: 5981/81503-A/GJG/ACK

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which:
(check one)

____ *is attached hereto.*

_X_ *was filed on* October 21, 2011 *as*

Application Serial No. 13/278,333

and was amended October 21, 2011
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

JPW Rev. 8/13/08

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Pending
61/406,067	October 22, 2010	Pending

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
N/A		

And I hereby appoint

Gary J. Gershik (Reg. No. 39,992); John P. White (Reg. No. 28,678); Christopher C. Dunham (Reg. No. 22,031); Norman H. Zivin (Reg. No. 25,385); William E. Pelton (Reg. No. 25,702); Robert D. Katz (Reg. No. 30,141); and Paul Teng (Reg. No. 40,837).

and each of them, all c/o Cooper & Dunham LLP, 30 Rockefeller Plaza, 20th Floor, New York, New York 10112, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to:

Gary J. Gershik, Esq. *Reg.No.* 39,992
Cooper & Dunham, LLP (Customer Number 23432)
30 Rockefeller Plaza
20th Floor
New York, New York 10112
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor James Hicks
Inventor's signature _____ *Date of signature* 11-16-11
Citizenship US
Residence 3 Great Meadow Road, Locust Valley, NY 11560
Post Office Address Same as above

Full name of additional joint inventor (if any) Nicholas Navin
Inventor's signature _____ *Date of signature* _____
Citizenship US
Residence 1621 Wichita St., Houston, TX 77004
Post Office Address Same as above

Full name of additional joint inventor (if any) Jennifer Troge
Inventor's signature _____ *Date of signature* 11.16.11
Citizenship US
Residence 100 Simcoe St., Oyster Bay, NY 11771
Post Office Address Same as above

GUARDANT HEALTH EX.1009.128

Full name of additional joint inventor (if any) Zihua Wang

Inventor's signature _____ *Date of signature* 11/16/11

Citizenship China

Residence 13 Meadowlawn St., Huntington, NY 11743

Post Office Address Same as above

Full name of additional joint inventor (if any) Michael Wigler

Inventor's signature _____ *Date of signature* 11/16/2011

Citizenship US

Residence 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Post Office Address Same as above

Full name of additional joint inventor (if any) _____

Inventor's signature _____ *Date of signature* _____

Citizenship _____

Residence _____

Post Office Address _____

Full name of additional joint inventor (if any) _____

Inventor's signature _____ *Date of signature* _____

Citizenship _____

Residence _____

Post Office Address _____

GUARDANT HEALTH EX.1009.129

Docket Number: 5981/81503-A/GJG/ACK

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which:
(check one)

____ *is attached hereto.*

_X_ *was filed on* October 21, 2011 *as*

Application Serial No. 13/278,333

and was amended October 21, 2011
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

JPW Rev. 8/13/08

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Pending
61/406,067	October 22, 2010	Pending

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
N/A		

And I hereby appoint

Gary J. Gershik (Reg. No. 39,992); John P. White (Reg. No. 28,678); Christopher C. Dunham (Reg. No. 22,031); Norman H. Zivin (Reg. No. 25,385); William E. Pelton (Reg. No. 25,702); Robert D. Katz (Reg. No. 30,141); and Paul Teng (Reg. No. 40,837).

and each of them, all c/o Cooper & Dunham LLP, 30 Rockefeller Plaza, 20th Floor, New York, New York 10112, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

GUARDANT HEALTH EX.1009.131

Please address all communications, and direct all telephone calls, regarding this application to:

Gary J. Gershik, Esq. *Reg.No.* 39,992
Cooper & Dunham, LLP (Customer Number 23432)
30 Rockefeller Plaza
20th Floor
New York, New York 10112
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor James Hicks

Inventor's signature _____ *Date of signature* _____

Citizenship US

Residence 3 Great Meadow Road, Locust Valley, NY 11560

Post Office Address Same as above

Full name of additional joint inventor (if any) Nicholas Navin

Inventor's signature Nicholas E Navin *Date of signature* 11-14-2011

Citizenship US

Residence 1621 Wichita St., Houston, TX 77004

Post Office Address Same as above

Full name of additional joint inventor (if any) Jennifer Troge

Inventor's signature _____ *Date of signature* _____

Citizenship US

Residence 100 Simcoe St., Oyster Bay, NY 11771

Post Office Address Same as above

GUARDANT HEALTH EX.1009.132

Full name of additional joint inventor (if any) Zihua Wang

Inventor's signature _____ *Date of signature* _____

Citizenship China

Residence 13 Meadowlawn St., Huntington, NY 11743

Post Office Address Same as above

Full name of additional joint inventor (if any) Michael Wigler

Inventor's signature _____ *Date of signature* _____

Citizenship US

Residence 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Post Office Address Same as above

Full name of additional joint inventor (if any) _____

Inventor's signature _____ *Date of signature* _____

Citizenship _____

Residence _____

Post Office Address _____

Full name of additional joint inventor (if any) _____

Inventor's signature _____ *Date of signature* _____

Citizenship _____

Residence _____

Post Office Address _____

GUARDANT HEALTH EX.1009.133

Attorney Docket No. 5981/81503-A/GIG/ACK

Assignment of Non-Provisional Application

In consideration of One Dollar ($1.00), and other good and valuable consideration, the receipt of which is hereby acknowledged, we, the undersigned,

James Hicks residing at 3 Great Meadow Road, Locust Valley, NY 11560; Nicholas Navin residing at 1621 Wichita St., Houston, TX 77004; Jennifer Troge residing at 100 Simcoe St., Oyster Bay, NY 11771; Zihua Wang residing at 13 Meadowlawn St., Huntington, NY 11743; and Michael Wigler residing at 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Hereby sell, assign and transfer *to* Cold Spring Harbor Laboratory, *a corporation of the State of* New York, *having a place of business at* One Bungtown Road, Cold Spring Harbor, New York 11724 U.S.A, *in the County of* Suffolk *and State of* New York, *its successors, assigns and legal representatives, the entire right, title and interest for all countries, in and to any and all inventions which are disclosed and claimed, and any and all inventions which are disclosed but not claimed, in the application for United States Patent, which has been executed by the undersigned on* November 16, 2011; November 14, 2011; November 16, 2011; November 16, 2011; and November 16, 2011 *and is entitled*

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

(U.S. Serial No. 13/278,333, filed October 21, 2011, claiming the benefit of U.S. Provisional Application Nos. 61/510,579, filed July 22, 2011 and 61/406,067, filed October 22, 2010)

and in and to said application and all divisional, continuing, substitute, renewal, reissue, and all other applications for U.S. Letters Patent or other related property rights in any and all foreign countries which have been or shall be filed on any of said inventions disclosed in said application; and in and to all original and reissued patents or related foreign documents which have been or shall be issued on said inventions;

Authorize and request *the Commissioner for Patents of the United States to issue to said Assignee, the corporation above named, its successors, assigns and legal representatives, in accordance with this assignment, any and all United States Letters Patent on said inventions or any of them disclosed in said application;*

GUARDANT HEALTH EX.1009.134

***Agree** that said Assignee may apply for and receive foreign Letters Patent or rights of any other kind for said inventions, or any of them; and may claim, in applications for said foreign Letters Patent or other rights, the priority of the aforesaid United States patent application under the provisions of the International Convention of 1883 and later modifications thereof, under the Patent Cooperation Treaty, under the European Patent Convention or under any other available international agreement; and that, when requested, without charge to, but at the expense of, said Assignee, its successors, assigns and legal representatives, to carry out in good faith the intent and purpose of this assignment, the undersigned or the undersigned's executors or administrators will, for the United States and all foreign countries, execute all divisional, continuing, substitute, renewal, reissue, and all other patent applications or other documents on any and all said inventions; execute all rightful oaths, assignments, powers of attorney and other papers; communicate to said Assignee, its successors, assigns and representatives, all facts known and documents available to the undersigned relating to said inventions and the history thereof; testify in all legal proceedings; and generally do everything possible which said Assignee, its successors, assigns or representatives shall consider desirable for aiding in securing, maintaining and enforcing proper patent protection for said inventions and for vesting title to said inventions and all applications for patents or related foreign rights and all patents on said inventions, in said Assignee, its successors, assigns and legal representatives; and*

***Covenant** with said Assignee, its successors, assigns and legal representatives that no assignment, grant, mortgage, license or other agreement affecting the rights and property herein conveyed has been made to others by the undersigned, and that full right to convey the same as herein expressed is possessed by the undersigned.*

James Hicks [L.S.]

Date: 11-16-11

Witness: _____ [signature]

Vladimir Dorokeff [printed name]

107 Cleveland Dr [address]

Centerport, NY 11721

GUARDANT HEALTH EX.1009.135

[signature] [LS]
Nicholas Navin

Date: 11-14-2011
Witness: [signature]
Jill Waters [printed name]
1515 Holcombe Blvd [address]
Houston TX 77030

[LS]
Jennifer Troge

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

[LS]
Zihua Wang

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

[LS]
Michael Wigler

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

[LS]

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

GUARDANT HEALTH EX.1009.136

_____ [LS]
Nicholas Navin

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

_____ [LS]
Jennifer Troge

Date: Nov 16, 2011
Witness: _____ [signature]
Vladimir Drozdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

_____ [LS]
Zihua Wang

Date: 11/16/2011
Witness: _____ [signature]
Vladimir Drozdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

_____ [LS]
Michael Wigler

Date: 11/16/2011
Witness: _____ [signature]
Vladimir Drozdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

_____ [LS]

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

GUARDANT HEALTH EX.1009.137

Electronic Patent Application Fee Transmittal

Application Number:	
Filing Date:	
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Andrew Gorman
Attorney Docket Number:	81503-AA/GJG/AWG

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Utility filing Fee (Electronic filing)	4011	1	70	70
Utility Search Fee	2111	1	300	300
Utility Examination Fee	2311	1	360	360
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Petition Fee-37CFR 1.17(h) (Group II)	2464	1	70	70

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				
Miscellaneous:				
		Total in USD ($)		**800**

Electronic Acknowledgement Receipt

EFS ID:	25127274
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Andrew Gorman
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG
Receipt Date:	07-MAR-2016
Filing Date:	
Time Stamp:	19:09:10
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	Deposit Account
Payment was successfully received in RAM	$800
RAM confirmation Number	6102
Deposit Account	033125
Authorized User	COOPER & DUNHAM LLP

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.140

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Transmittal of New Application	160307_81503_AA_Application_Transmittal_AC.pdf	134763 b49383fe4befa2bc0ad3214be60e424fe26938b2	no	3
Warnings:					
Information:					
2		160307_81503_AA_Preliminary_Amendment_AC.pdf	454341 3454dcaf55366bc4469333e85c61271b86c404a0	yes	11

Multipart Description/PDF files in .zip description		
Document Description	**Start**	**End**
Preliminary Amendment	1	1
Claims	2	8
Applicant Arguments/Remarks Made in an Amendment	9	11

Warnings:					
Information:					
3	Application Data Sheet	160307_81503_AA_Application_Data_Sheet_AC.pdf	648033 0a0190c7e6ff8d36b127a6a852fa8e0f952f978e	no	10
Warnings:					
Information:					
This is not an USPTO supplied ADS fillable form					
4		160307_81503_AA_Specification_AC.pdf	4178776 11f965a94663eed2987a0369dfb94ffefccbecf1	yes	87

Multipart Description/PDF files in .zip description		
Document Description	**Start**	**End**
Specification	1	69
Claims	70	86
Abstract	87	87

Warnings:					
Information:					
5	Drawings-other than black and white line drawings	160307_81503_AA_Drawings_Color_AC.pdf	1022303 cba5a78dc100204a4a55c0796e2a310dd89f45b6	no	11
Warnings:					
The page size in the PDF is too large. The pages should be 8.5 x 11 or A4. If this PDF is submitted, the pages will be resized upon entry into the Image File Wrapper and may affect subsequent processing					
Information:					
6	Petition for review by the Office of Petitions	160307_81503_AA_Petition_Color_Drawings_AC.pdf	107631 5af8d36a09da59425ef99e5254ee6abe808479f4	no	3
Warnings:					
Information:					
7	Oath or Declaration filed	160307_81503_AA_Declaration_AC.pdf	279560 2de6a84a7a599ed2c6b9552b10822792f741a305	no	8
Warnings:					
Information:					
8	Assignee showing of ownership per 37 CFR 3.73	160307_81503_AA_Assignment_AC.pdf	163073 fd658f09df89b842adb2b58f88ac0eb5ca8abe42	no	4
Warnings:					
Information:					
9	Fee Worksheet (SB06)	fee-info.pdf	37378 935b6868da9fd3e0aa606be97c2b1acf2a2c9561	no	2
Warnings:					
Information:					
			Total Files Size (in bytes):	7025858	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

DocCode – SCORE

SCORE Placeholder Sheet for IFW Content

Application Number: 15063278 Document Date: 03/07/2016

The presence of this form in the IFW record indicates that the following document type was received in electronic format on the date identified above. This content is stored in the SCORE database.

Since this was an electronic submission, there is no physical artifact folder, no artifact folder is recorded in PALM, and no paper documents or physical media exist. The TIFF images in the IFW record were created from the original documents that are stored in SCORE.

- Drawing

At the time of document entry (noted above):

- USPTO employees may access SCORE content via eDAN using the Supplemental Content tab, or via the SCORE web page.
- External customers may access SCORE content via PAIR using the Supplemental Content tab.

Form Revision Date: August 26, 2013

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : James Hicks et al.

U.S. Serial No.: 15/063,278

Filed : March 7, 2016

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
20th Floor
New York, New York 10112
March 8, 2016

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

Sir:

SUPPLEMENTAL PRELIMINARY AMENDMENT

This Preliminary Amendment is submitted in connection with the above-identified application prior to the mailing of a first Office Action.

Please amend the subject application as follows:

Amendments to Specification begin on page **2** of this paper; and

Remarks begin on page **3** of this paper.

Amendments to the Specification

Please amend the paragraph beginning on page 1, lines 4-7, as follows:

--This application is a continuation of U.S. Serial No. 13/278,333, filed October 21, 2011, now allowed, which claims the benefit of U.S. Provisional Application Nos. 61/510,579, filed July 22, 2011, and 61/406,067, filed October 22, 2010 the contents of each of which are hereby incorporated by reference in their entirety into this application.--

REMARKS

The subject application is a continuation of co-pending U.S Serial No. 13/278,333, filed October 21, 2011. By this Preliminary Amendment, applicants have amended the specification to recite the continuing data for the above-identified parent application.

Applicants maintain that the amendments to the specification raise no issue of new matter. Accordingly, applicants respectfully request entry of this Supplemental Preliminary Amendment.

If a telephone conference would be of assistance in advancing prosecution of this application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, is deemed necessary in connection with the filing of this Preliminary Amendment and the accompanying application. However, if any additional fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmittal

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

Andrew W. Gorman 3/8/16
Date

Electronic Acknowledgement Receipt

EFS ID:	25137272
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Joseph Micali
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG
Receipt Date:	08-MAR-2016
Filing Date:	
Time Stamp:	16:07:25
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Preliminary Amendment	160308_5981_81503_AA_Supplemental_Preliminary_Amendment_AWG.pdf	96310 9f6530ffae106e0f8b9a4abeb1746fdb3185f35d	no	4

Warnings:	
Information:	

GUARDANT HEALTH EX.1009.149

Total Files Size (in bytes):	96310

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING or 371(c) DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	TOT CLAIMS	IND CLAIMS
15/063,278	03/07/2016	1634	730	81503-AA/GJG/AWG/AC	20	2

CONFIRMATION NO. 4038

FILING RECEIPT

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

OC000000081608931

Date Mailed: 03/22/2016

Receipt is acknowledged of this non-provisional patent application. The application will be taken up for examination in due course. Applicant will be notified as to the results of the examination. Any correspondence concerning the application must include the following identification information: the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. **If an error is noted on this Filing Receipt, please submit a written request for a Filing Receipt Correction. Please provide a copy of this Filing Receipt with the changes noted thereon. If you received a "Notice to File Missing Parts" for this application, please submit any corrections to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections**

Inventor(s)

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

Applicant(s)

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

Assignment For Published Patent Application

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY

Power of Attorney:

Christopher Dunham--22031
Norman Zivin--25385
William Pelton--25702
John White--28678
Robert Katz--30141
Gary Gershik--39992
Paul Teng--40837

Domestic Priority data as claimed by applicant

This application is a CON of 13/278,333 10/21/2011
which claims benefit of 61/510,579 07/22/2011
and claims benefit of 61/406,067 10/22/2010

GUARDANT HEALTH EX.1009.151

Foreign Applications for which priority is claimed (You may be eligible to benefit from the **Patent Prosecution Highway** program at the USPTO. Please see http://www.uspto.gov for more information.) - None.
Foreign application information must be provided in an Application Data Sheet in order to constitute a claim to foreign priority. See 37 CFR 1.55 and 1.76.

Permission to Access Application via Priority Document Exchange: Yes

Permission to Access Search Results: Yes

Applicant may provide or rescind an authorization for access using Form PTO/SB/39 or Form PTO/SB/69 as appropriate.

If Required, Foreign Filing License Granted: 03/22/2016

The country code and number of your priority application, to be used for filing abroad under the Paris Convention, is **US 15/063,278**

Projected Publication Date: To Be Determined - pending completion of Sequence Disclosures

Non-Publication Request: No

Early Publication Request: No
**** SMALL ENTITY ****
Title

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Preliminary Class

435

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications: No

PROTECTING YOUR INVENTION OUTSIDE THE UNITED STATES

Since the rights granted by a U.S. patent extend only throughout the territory of the United States and have no effect in a foreign country, an inventor who wishes patent protection in another country must apply for a patent in a specific country or in regional patent offices. Applicants may wish to consider the filing of an international application under the Patent Cooperation Treaty (PCT). An international (PCT) application generally has the same effect as a regular national patent application in each PCT-member country. The PCT process **simplifies** the filing of patent applications on the same invention in member countries, but **does not result** in a grant of "an international patent" and does not eliminate the need of applicants to file additional documents and fees in countries where patent protection is desired.

Almost every country has its own patent law, and a person desiring a patent in a particular country must make an application for patent in that country in accordance with its particular laws. Since the laws of many countries differ in various respects from the patent law of the United States, applicants are advised to seek guidance from specific foreign countries to ensure that patent rights are not lost prematurely.

Applicants also are advised that in the case of inventions made in the United States, the Director of the USPTO must issue a license before applicants can apply for a patent in a foreign country. The filing of a U.S. patent application

GUARDANT HEALTH EX.1009.152

serves as a request for a foreign filing license. The application's filing receipt contains further information and guidance as to the status of applicant's license for foreign filing.

Applicants may wish to consult the USPTO booklet, "General Information Concerning Patents" (specifically, the section entitled "Treaties and Foreign Patents") for more information on timeframes and deadlines for filing foreign patent applications. The guide is available either by contacting the USPTO Contact Center at 800-786-9199, or it can be viewed on the USPTO website at http://www.uspto.gov/web/offices/pac/doc/general/index.html.

For information on preventing theft of your intellectual property (patents, trademarks and copyrights), you may wish to consult the U.S. Government website, http://www.stopfakes.gov. Part of a Department of Commerce initiative, this website includes self-help "toolkits" giving innovators guidance on how to protect intellectual property in specific countries such as China, Korea and Mexico. For questions regarding patent enforcement issues, applicants may call the U.S. Government hotline at 1-866-999-HALT (1-866-999-4258).

LICENSE FOR FOREIGN FILING UNDER

Title 35, United States Code, Section 184

Title 37, Code of Federal Regulations, 5.11 & 5.15

GRANTED

The applicant has been granted a license under 35 U.S.C. 184, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" followed by a date appears on this form. Such licenses are issued in all applications where the conditions for issuance of a license have been met, regardless of whether or not a license may be required as set forth in 37 CFR 5.15. The scope and limitations of this license are set forth in 37 CFR 5.15(a) unless an earlier license has been issued under 37 CFR 5.15(b). The license is subject to revocation upon written notification. The date indicated is the effective date of the license, unless an earlier license of similar scope has been granted under 37 CFR 5.13 or 5.14.

This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

The grant of a license does not in any way lessen the responsibility of a licensee for the security of the subject matter as imposed by any Government contract or the provisions of existing laws relating to espionage and the national security or the export of technical data. Licensees should apprise themselves of current regulations especially with respect to certain countries, of other agencies, particularly the Office of Defense Trade Controls, Department of State (with respect to Arms, Munitions and Implements of War (22 CFR 121-128)); the Bureau of Industry and Security, Department of Commerce (15 CFR parts 730-774); the Office of Foreign AssetsControl, Department of Treasury (31 CFR Parts 500+) and the Department of Energy.

NOT GRANTED

No license under 35 U.S.C. 184 has been granted at this time, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" DOES NOT appear on this form. Applicant may still petition for a license under 37 CFR 5.12, if a license is desired before the expiration of 6 months from the filing date of the application. If 6 months has lapsed from the filing date of this application and the licensee has not received any indication of a secrecy order under 35 U.S.C. 181, the licensee may foreign file the application pursuant to 37 CFR 5.15(b).

GUARDANT HEALTH EX.1009.153

SelectUSA

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The U.S. offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to promote and facilitate business investment. SelectUSA provides information assistance to the international investor community; serves as an ombudsman for existing and potential investors; advocates on behalf of U.S. cities, states, and regions competing for global investment; and counsels U.S. economic development organizations on investment attraction best practices. To learn more about why the United States is the best country in the world to develop technology, manufacture products, deliver services, and grow your business, visit http://www.SelectUSA.gov or call +1-202-482-6800.

GUARDANT HEALTH EX.1009.154

PATENT APPLICATION FEE DETERMINATION RECORD

Substitute for Form PTO-875

Application or Docket Number: 15/063,278

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA	SMALL ENTITY RATE($)	SMALL ENTITY FEE($)	OR	OTHER THAN SMALL ENTITY RATE($)	OTHER THAN SMALL ENTITY FEE($)
BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A	70		N/A	
SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A	300		N/A	
EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A	360		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	20 minus 20 =	*	x 40 =	0.00	OR		
INDEPENDENT CLAIMS (37 CFR 1.16(h))	2 minus 3 =	*	x 210 =	0.00			
APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).			0.00			
MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))				0.00			
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL	730		TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	SMALL ENTITY RATE($)	SMALL ENTITY ADDITIONAL FEE($)	OR	OTHER THAN SMALL ENTITY RATE($)	OTHER THAN SMALL ENTITY ADDITIONAL FEE($)
AMENDMENT A	Total (37 CFR 1.16(i))	*	Minus	**	=	x =		OR	x =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	x =		OR	x =	
	Application Size Fee (37 CFR 1.16(s))									
	FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE		OR	TOTAL ADD'L FEE	

		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE($)	ADDITIONAL FEE($)		RATE($)	ADDITIONAL FEE($)
AMENDMENT B	Total (37 CFR 1.16(i))	*	Minus	**	=	x =		OR	x =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	x =		OR	x =	
	Application Size Fee (37 CFR 1.16(s))									
	FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE		OR	TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".
The "Highest Number Previously Paid For" (Total or Independent) is the highest found in the appropriate box in column 1.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

FORMALITIES LETTER

OC000000081608932

Date Mailed: 03/22/2016

NOTICE TO COMPLY WITH REQUIREMENTS FOR PATENT APPLICATIONS CONTAINING NUCLEOTIDE SEQUENCE AND/OR AMINO ACID SEQUENCE DISCLOSURES

Filing Date Granted

Applicant is given **TWO MONTHS FROM THE DATE OF THIS NOTICE** within which to comply with the sequence rules, 37 CFR §§ 1.821-1.825. Failure to comply with these requirements will result in ABANDONMENT of the application under 37 CFR § 1.821(g). Extension of time may be obtained by filing a petition accompanied by the extension fee under the provisions of 37 CFR § 1.136.

- This application does not contain, as a separate part of the disclosure on paper copy, a "Sequence Listing" as required by 37 CFR 1.821(c) Applicant must provide an initial paper or compact disc copy of the "Sequence Listing", **as well as an amendment specifically directing its entry into the application** and a statement that the content of the sequence listing information recorded in computer readable form is identical to the written (on paper or compact disc) sequence listing and, where applicable, includes no new matter, as required by 37 CFR 1.821(e), 1.821(f), 1.821(g), 1.825(b), or 1.825(d). If the effective filing date is on or after September 8, 2000, see the final rulemaking notice published in the Federal Register at 65 FR 54604 (September 8, 2000) and 1238 OG 145 (September 19, 2000).
- A copy of the "Sequence Listing" in computer readable form has not been submitted as required by 37 CFR 1.821(e). If the effective filing date is on or after September 8, 2000, see the final rulemaking notice published in the Federal Register at 65 FR 54604 (September 8, 2000) and 1238 OG 145 (September 19, 2000). Applicant must provide an initial computer readable form (CRF) copy of the "Sequence Listing" and a statement that the content of the sequence listing information recorded in computer readable form is identical to the written (on paper or compact disc) sequence listing and, where applicable, includes no new matter, as required by 37 CFR 1.821(e), 1.821(f), 1.821(g), 1.825(b), or 1.825(d). If applicant desires the sequence listing in the instant application to be identical with that of another application on file in the U.S. Patent and Trademark Office, such request in accordance with 37 CFR 1.821(e) may be submitted in lieu of a new CRF.

Applicant is cautioned that correction of the above items may cause the specification and drawings page count to exceed 100 pages. If the specification and drawings exceed 100 pages, applicant will need to submit the required application size fee.

For questions regarding compliance to these requirements, please contact:

- **For Rules interpretation, call (571) 272-2510**
- **For CRF submission help, call (571) 272-2510**
- **For PatentIn software program support technical assistance, call (866)217-9197.**

GUARDANT HEALTH EX.1009.156

- **Send e-mail correspondence for PatentIn software program help to ebc@uspto.gov**
- **Patent software is available at http://www.uspto.gov/patents/resources/tools/checker/patentinrel.jsp .**

Items Required To Avoid Processing Delays:

Applicant is notified that the above-identified application contains the deficiencies noted below. No period for reply is set forth in this notice for correction of these deficiencies. However, if a deficiency relates to the inventor's oath or declaration, the applicant must file an oath or declaration in compliance with 37 CFR 1.63, or a substitute statement in compliance with 37 CFR 1.64, executed by or with respect to each actual inventor no later than the expiration of the time period set in the "Notice of Allowability" to avoid abandonment. See 37 CFR 1.53(f).

A new inventor's oath or declaration that identifies this application (e.g., by Application Number and filing date) is required. The inventor's oath or declaration does not comply with 37 CFR 1.63 in that it:

- does not state that the above-identified application was made or authorized to be made by the person executing the oath or declaration.

James Hicks
Nicholas Navin
Jennifer Troge
Zihua Wang
Michael Wigler

Replies must be received in the USPTO within the set time period or must include a proper Certificate of Mailing or Transmission under 37 CFR 1.8 with a mailing or transmission date within the set time period. For more information and a suggested format, see Form PTO/SB/92 and MPEP 512.

Replies should be mailed to:

Mail Stop Missing Parts
Commissioner for Patents
P.O. Box 1450
Alexandria VA 22313-1450

Registered users of EFS-Web may alternatively submit their reply to this notice via EFS-Web, including a copy of this Notice and selecting the document description "Applicant response to Pre-Exam Formalities Notice". https://sportal.uspto.gov/authenticate/AuthenticateUserLocalEPF.html

For more information about EFS-Web please call the USPTO Electronic Business Center at 1-866-217-9197 or visit our website at http://www.uspto.gov/ebc.

If you are not using EFS-Web to submit your reply, you must include a copy of this notice.

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at **(571) 272-4000** or **(571) 272-4200** or **1-888-786-0101**.

/tpetros/

GUARDANT HEALTH EX.1009.157

DocCode – SEQ.TXT

SCORE Placeholder Sheet for IFW Content

Application Number: 15063278 Document Date: 05/20/2016

The presence of this form in the IFW record indicates that the following document type was received in electronic format on the date identified above. This content is stored in the SCORE database.

Since this was an electronic submission, there is no physical artifact folder, no artifact folder is recorded in PALM, and no paper documents or physical media exist. The TIFF images in the IFW record were created from the original documents that are stored in SCORE.

- Sequence Listing

At the time of document entry (noted above):

- USPTO employees may access SCORE content via eDAN using the Supplemental Content tab, or via the SCORE web page.
- External customers may access SCORE content via PAIR using the Supplemental Content tab.

Form Revision Date: August 26, 2013

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : James Hicks et al.

U.S. Serial No. : 15/063,278

Filed : March 7, 2016

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
20th Floor
New York, New York 10112
May 20, 2016

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

AMENDMENT IN RESPONSE TO MARCH 22, 2016 NOTICE TO COMPLY WITH REQUIREMENTS FOR PATENT APPLICATIONS CONTAINING NUCLEOTIDE SEQUENCE AND/OR AMINO ACID SEQUENCE DISCLOSURES – FILING DATE GRANTED

This Amendment is submitted in response to the March 22, 2016 Notice To Comply With Requirements For Patent Applications Containing Nucleotide Sequence And/Or Amino Acid Sequence Disclosures - Filing Date Granted (hereinafter "March 22, 2016 Notice"), which issued in connection with the above-identified application. A copy of the March 22, 2016 Notice is attached hereto as **Exhibit A.**

Amendments to the Specification begin on page 2 of this paper.

A **Sequence Listing** is referred to on page 3 of this paper and filed herewith as a **text file.**

A **Statement of Compliance under 37 C.F.R. § 1.825(a)** is referred to on page 4 of this paper and attached hereto as **Exhibit B.**

Remarks begin on page 4 of this paper.

Amendment to the Specification

Please insert the following new section heading and paragraph on page 1, immediately before the section titled, "Background of the Invention":

Reference to a Sequence Listing

This application incorporates-by-reference nucleotide and/or amino acid sequences which are present in the file named "160520_81503_AA_Sequence_Listing_JJC.txt" which is 2.59 kilobytes in size, and which was created May 13, 2016 in the IBM-PC machine format, having an operating system compatibility with MS-Windows, which is contained in the text file filed May 20, 2016 as part of this application.

Sequence Listing

Please amend the subject application to include the Sequence Listing filed herewith as a text file named "160520_81503_AA_Sequence_Listing_JJC.txt".

Remarks

Amendment to the Specification

By this amendment, applicants have amended the specification to insert a section heading and paragraph indicating the name, size, date of creation, and date of filing of the text file named "160520_81503_AA_Sequence_Listing_JJC.txt" which corresponds to the Sequence Listing.

Applicants maintain that the amendments made hereinabove do not raise any issue of new matter. Accordingly, applicants respectfully request entry of this Amendment.

Sequence Listing

Applicants attach hereto as **Exhibit B** a Statement of Compliance under 37 C.F.R. § 1.825(a) certifying that the Sequence Listing submitted herewith as a text file does not introduce any new matter into the subject application.

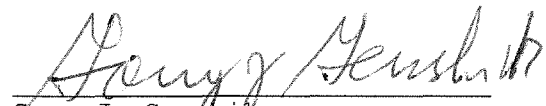
The subject matter in the Sequence Listing submitted herewith corresponds to the subject matter in Figures 8 and 9 of the subject application as filed.

In accordance with the guidance set forth in section I1 of "Legal Framework for EFS-Web (06April11)" published by the U.S. Patent and Trademark Office at www.uspto.gov/patents/process/file/efs/guidance/New_legal_framework.jsp, applicants are not submitting herewith a statement under 37 C.F.R. § 1.821(f), since the text file submitted herewith will serve as both the "paper copy" and the "computer readable form" of the Sequence Listing.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

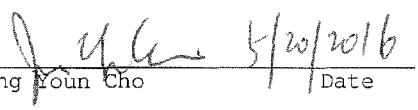
No fee is deemed necessary in connection with the filing of this Amendment. However, if any additional fee is required, authorization is hereby given to charge the amount of any such additional fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorney for Applicant
Cooper & Dunham LLP (Customer #23432)
30 Rockefeller Plaza, 20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.



Joong Youn Cho 5/20/2016
Date

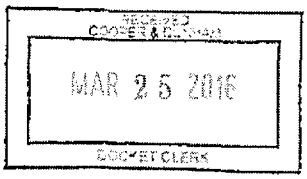
UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112



FORMALITIES LETTER



Date Mailed: 03/22/2016

NOTICE TO COMPLY WITH REQUIREMENTS FOR PATENT APPLICATIONS CONTAINING NUCLEOTIDE SEQUENCE AND/OR AMINO ACID SEQUENCE DISCLOSURES

Filing Date Granted

Applicant is given **TWO MONTHS FROM THE DATE OF THIS NOTICE** within which to comply with the sequence rules, 37 CFR §§ 1.821-1.825. Failure to comply with these requirements will result in ABANDONMENT of the application under 37 CFR § 1.821(g). Extension of time may be obtained by filing a petition accompanied by the extension fee under the provisions of 37 CFR § 1.136.

- This application does not contain, as a separate part of the disclosure on paper copy, a "Sequence Listing" as required by 37 CFR 1.821(c) Applicant must provide an initial paper or compact disc copy of the "Sequence Listing", **as well as an amendment specifically directing its entry into the application** and a statement that the content of the sequence listing information recorded in computer readable form is identical to the written (on paper or compact disc) sequence listing and, where applicable, includes no new matter, as required by 37 CFR 1.821(e), 1.821(f), 1.821(g), 1.825(b), or 1.825(d). If the effective filing date is on or after September 8, 2000, see the final rulemaking notice published in the Federal Register at 65 FR 54604 (September 8, 2000) and 1238 OG 145 (September 19, 2000).
- A copy of the "Sequence Listing" in computer readable form has not been submitted as required by 37 CFR 1.821(e). If the effective filing date is on or after September 8, 2000, see the final rulemaking notice published in the Federal Register at 65 FR 54604 (September 8, 2000) and 1238 OG 145 (September 19, 2000). Applicant must provide an initial computer readable form (CRF) copy of the "Sequence Listing" and a statement that the content of the sequence listing information recorded in computer readable form is identical to the written (on paper or compact disc) sequence listing and, where applicable, includes no new matter, as required by 37 CFR 1.821(e), 1.821(f), 1.821(g), 1.825(b), or 1.825(d). If applicant desires the sequence listing in the instant application to be identical with that of another application on file in the U.S. Patent and Trademark Office, such request in accordance with 37 CFR 1.821(e) may be submitted in lieu of a new CRF.

Applicant is cautioned that correction of the above items may cause the specification and drawings page count to exceed 100 pages. If the specification and drawings exceed 100 pages, applicant will need to submit the required application size fee.

For questions regarding compliance to these requirements, please contact:

- **For Rules interpretation, call (571) 272-2510**
- **For CRF submission help, call (571) 272-2510**
- **For PatentIn software program support technical assistance, call (866)217-9197.**

Applicant: Hicks et al.
Serial No.: 15/063,278
Filed: March 7, 2016
Exhibit A

GUARDANT HEALTH EX.1009.164

- **Send e-mail correspondence for PatentIn software program help to ebc@uspto.gov**
- **Patent software is available at http://www.uspto.gov/patents/resources/tools/checker/patentinrel.jsp .**

Items Required To Avoid Processing Delays:

Applicant is notified that the above-identified application contains the deficiencies noted below. No period for reply is set forth in this notice for correction of these deficiencies. However, if a deficiency relates to the inventor's oath or declaration, the applicant must file an oath or declaration in compliance with 37 CFR 1.63, or a substitute statement in compliance with 37 CFR 1.64, executed by or with respect to each actual inventor no later than the expiration of the time period set in the "Notice of Allowability" to avoid abandonment. See 37 CFR 1.53(f).

A new inventor's oath or declaration that identifies this application (e.g., by Application Number and filing date) is required. The inventor's oath or declaration does not comply with 37 CFR 1.63 in that it:

- does not state that the above-identified application was made or authorized to be made by the person executing the oath or declaration.

James Hicks
Nicholas Navin
Jennifer Troge
Zihua Wang
Michael Wigler

Replies must be received in the USPTO within the set time period or must include a proper Certificate of Mailing or Transmission under 37 CFR 1.8 with a mailing or transmission date within the set time period. For more information and a suggested format, see Form PTO/SB/92 and MPEP 512.

Replies should be mailed to:

Mail Stop Missing Parts
Commissioner for Patents
P.O. Box 1450
Alexandria VA 22313-1450

Registered users of EFS-Web may alternatively submit their reply to this notice via EFS-Web, including a copy of this Notice and selecting the document description "Applicant response to Pre-Exam Formalities Notice". https://sportal.uspto.gov/authenticate/AuthenticateUserLocalEPF.html

For more information about EFS-Web please call the USPTO Electronic Business Center at 1-866-217-9197 or visit our website at http://www.uspto.gov/ebc.

If you are not using EFS-Web to submit your reply, you must include a copy of this notice.

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at (571) 272-4000 or (571) 272-4200 or 1-888-786-0101.

/tpetros/

GUARDANT HEALTH EX.1009.165

Docket No. 81503-AA/GJG/JJC

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.
U.S. Serial No.	:	15/063,278
Filed	:	March 7, 2016
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
20th Floor
New York, New York 10112
May 20, 2016

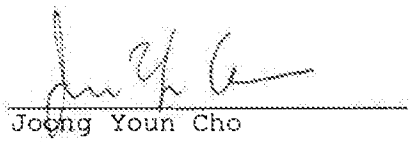
BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

STATEMENT OF COMPLIANCE UNDER 37 C.F.R. § 1.821(e)
IN CONNECTION WITH ABOVE-IDENTIFIED APPLICATION

Pursuant to 37 C.F.R. § 1.821(e), I hereby certify that the content of the Sequence Listing submitted herewith as a text file named "160520_81503_AA_Sequence_Listing_JJC" is identical to the written sequence listing and does not introduce any new matter into the subject application.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully submitted,



Joong Youn Cho
c/o Cooper & Dunham LLP
30 Rockefeller Plaza, 20th Fl.
New York, New York 10112
Attn.: Gary J. Gershik
(212) 278-0400

Applicant: Hicks et al.
Serial No.: 15/063,278
Filed: March 7, 2016
Exhibit B

Electronic Acknowledgement Receipt

EFS ID:	25844035
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Joong Youn Cho
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	20-MAY-2016
Filing Date:	07-MAR-2016
Time Stamp:	17:14:17
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		160520_81503_AA_Amendment__JJC.pdf	493988 6d2e864e304ecd8b86dcb55fc2dc99e5a5eeffd2	yes	8

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Response to Pre-Exam Sequence Notice	1	1
	Specification	2	2
	Sequence Listing	3	3
	Applicant Arguments/Remarks Made in an Amendment	4	4
	Response to Pre-Exam Sequence Notice	5	7
	CRF Statement Paper and CRF are the same	8	8

Warnings:

The page size in the PDF is too large. The pages should be 8.5 x 11 or A4. If this PDF is submitted, the pages will be resized upon entry into the Image File Wrapper and may affect subsequent processing

Information:

2	Sequence Listing (Text File)	160520_81503_AA_Sequence_Listing_JJC.txt	2658	no	0

Warnings:

Information:

Total Files Size (in bytes):	496646

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

==========
Sequence Listing was accepted.
If you need help call the Patent Electronic Business Center at (866) 217-9197 (toll free).
Reviewer: Saleem, Syed (ASRC)
Timestamp: [year=2016; month=5; day=24; hr=8; min=14; sec=40; ms=54;]
==========

Validated By CRFValidator v 1.0.5

Application No: 15063278 Version No: 1.0

Input Set:

Output Set:

Started: 2016-05-20 17:14:21.724
Finished: 2016-05-20 17:14:23.392
Elapsed: 0 hr(s) 0 min(s) 1 sec(s) 668 ms
Total Warnings: 18
Total Errors: 0
No. of SeqIDs Defined: 8
Actual SeqID Count: 8

Error code	Error Description
W 213	Artificial or Unknown found in <213> in SEQ ID (1)
W 213	Artificial or Unknown found in <213> in SEQ ID (2)
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 447	n or Xaa used, for: SEQID(2) on line number 59
W 213	Artificial or Unknown found in <213> in SEQ ID (3)
W 213	Artificial or Unknown found in <213> in SEQ ID (4)
W 213	Artificial or Unknown found in <213> in SEQ ID (5)
W 213	Artificial or Unknown found in <213> in SEQ ID (6)
W 213	Artificial or Unknown found in <213> in SEQ ID (7)
W 213	Artificial or Unknown found in <213> in SEQ ID (8)

SEQUENCE LISTING

<110> Hicks, James
Navin, Nicholas
Troge, Jennifer
Wang, Zihua
Wigler, Michael

<120> VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

<130> 81503-AA/GJG/AWG/AC

<140> US 15/063,278
<141> 2016-03-07

<150> 13/278,333
<151> 2011-10-21

<150> 61406067
<151> 2010-10-22

<150> 61510579
<151> 2011-07-22

<160> 8

<170> PatentIn version 3.5

<210> 1
<211> 15
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of 3' end of nucleotide sequence after Terminal Transferase step

<400> 1
catgaaaaaa aaaaa 15

<210> 2
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence comprising a sequence complementary to portion of 3' end of a nucleotide sequence, and a varietal tag

<220>
<221> misc_feature
<222> (1)..(10)
<223> n is a, c, g, or t

<400> 2
nnnnnnnnnn ttttttttt catg 24

<210> 3
<211> 11
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of 3' end of nuceotide sequence after Pool, Cleanup, Melt, Terminal Transferase

<400> 3
ccccccccc c 11

<210> 4
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of a primer complementary to portion of 3' end of nucleotide sequence

<400> 4
gggggggggg 10

<210> 5
<211> 12
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of 3' end of nucleotide sequence after Terminal Transferase step

<400> 5
catgaaaaaa aa 12

<210> 6
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of 3' end of nucleotide sequence after Terminal Transferase step

<400> 6
catgaaaaaa aaaaaa 16

<210> 7

<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of primer complementary to portion of 3' end of nucleotide sequence

<400> 7
tttttttttt ttcatg 16

<210> 8
<211> 14
<212> DNA
<213> Artificial Sequence

<220>
<223> Portion of 3' end of nucleotide segment after Terminal Transferase step

<400> 8
catgaaaaaa aaaa 14

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

INFORMAL NOTICE

OC000000083175223

Date Mailed: 05/25/2016

INFORMATIONAL NOTICE TO APPLICANT

Applicant is notified that the above-identified application contains the deficiencies noted below. No period for reply is set forth in this notice for correction of these deficiencies. However, if a deficiency relates to the inventor's oath or declaration, the applicant must file an oath or declaration in compliance with 37 CFR 1.63, or a substitute statement in compliance with 37 CFR 1.64, executed by or with respect to each actual inventor no later than the expiration of the time period set in the "Notice of Allowability" to avoid abandonment. See 37 CFR 1.53(f).

The item(s) indicated below are also required and should be submitted with any reply to this notice to avoid further processing delays.

A new inventor's oath or declaration that identifies this application (e.g., by Application Number and filing date) is required. The inventor's oath or declaration does not comply with 37 CFR 1.63 in that it:

- does not state that the above-identified application was made or authorized to be made by the person executing the oath or declaration.

James Hicks
Nicholas Navin
Jennifer Troge
Zihua Wang
Michael Wigler

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at **(571) 272-4000** or **(571) 272-4200** or **1-888-786-0101**.

/ngfissha/

GUARDANT HEALTH EX.1009.175

PATENT APPLICATION FEE DETERMINATION RECORD

Substitute for Form PTO-875

Application or Docket Number: 15/063,278

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA	SMALL ENTITY RATE($)	SMALL ENTITY FEE($)	OR	OTHER THAN SMALL ENTITY RATE($)	OTHER THAN SMALL ENTITY FEE($)
BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A	70		N/A	
SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A	300		N/A	
EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A	360		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	20 minus 20 =	*	x 40 =	0.00	OR		
INDEPENDENT CLAIMS (37 CFR 1.16(h))	2 minus 3 =	*	x 210 =	0.00			
APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).			0.00			
MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))				0.00			
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL	730		TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	SMALL ENTITY RATE($)	SMALL ENTITY ADDITIONAL FEE($)	OR	OTHER THAN SMALL ENTITY RATE($)	OTHER THAN SMALL ENTITY ADDITIONAL FEE($)
AMENDMENT A	Total (37 CFR 1.16(i))	*	Minus	**	=	x =		OR	x =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	x =		OR	x =	
	Application Size Fee (37 CFR 1.16(s))									
	FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE		OR	TOTAL ADD'L FEE	

		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE($)	ADDITIONAL FEE($)		RATE($)	ADDITIONAL FEE($)
AMENDMENT B	Total (37 CFR 1.16(i))	*	Minus	**	=	x =		OR	x =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	x =		OR	x =	
	Application Size Fee (37 CFR 1.16(s))									
	FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							OR		
						TOTAL ADD'L FEE		OR	TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".
The "Highest Number Previously Paid For" (Total or Independent) is the highest found in the appropriate box in column 1.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING or 371(c) DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	TOT CLAIMS	IND CLAIMS
15/063,278	03/07/2016	1634	730	81503-AA/GJG/AWG/AC	20	2

CONFIRMATION NO. 4038

UPDATED FILING RECEIPT

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

OC000000083175222

Date Mailed: 05/25/2016

Receipt is acknowledged of this non-provisional patent application. The application will be taken up for examination in due course. Applicant will be notified as to the results of the examination. Any correspondence concerning the application must include the following identification information: the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. **If an error is noted on this Filing Receipt, please submit a written request for a Filing Receipt Correction. Please provide a copy of this Filing Receipt with the changes noted thereon. If you received a "Notice to File Missing Parts" for this application, please submit any corrections to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections**

Inventor(s)

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

Applicant(s)

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

Assignment For Published Patent Application

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY

Power of Attorney:

Christopher Dunham--22031
Norman Zivin--25385
William Pelton--25702
John White--28678
Robert Katz--30141
Gary Gershik--39992
Paul Teng--40837

Domestic Priority data as claimed by applicant

This application is a CON of 13/278,333 10/21/2011
which claims benefit of 61/510,579 07/22/2011
and claims benefit of 61/406,067 10/22/2010

GUARDANT HEALTH EX.1009.177

Foreign Applications for which priority is claimed (You may be eligible to benefit from the **Patent Prosecution Highway** program at the USPTO. Please see http://www.uspto.gov for more information.) - None.
Foreign application information must be provided in an Application Data Sheet in order to constitute a claim to foreign priority. See 37 CFR 1.55 and 1.76.

Permission to Access Application via Priority Document Exchange: Yes

Permission to Access Search Results: Yes

Applicant may provide or rescind an authorization for access using Form PTO/SB/39 or Form PTO/SB/69 as appropriate.

If Required, Foreign Filing License Granted: 03/22/2016

The country code and number of your priority application, to be used for filing abroad under the Paris Convention, is **US 15/063,278**

Projected Publication Date: 09/01/2016

Non-Publication Request: No

Early Publication Request: No
**** SMALL ENTITY ****
Title

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Preliminary Class

435

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications: No

PROTECTING YOUR INVENTION OUTSIDE THE UNITED STATES

Since the rights granted by a U.S. patent extend only throughout the territory of the United States and have no effect in a foreign country, an inventor who wishes patent protection in another country must apply for a patent in a specific country or in regional patent offices. Applicants may wish to consider the filing of an international application under the Patent Cooperation Treaty (PCT). An international (PCT) application generally has the same effect as a regular national patent application in each PCT-member country. The PCT process **simplifies** the filing of patent applications on the same invention in member countries, but **does not result** in a grant of "an international patent" and does not eliminate the need of applicants to file additional documents and fees in countries where patent protection is desired.

Almost every country has its own patent law, and a person desiring a patent in a particular country must make an application for patent in that country in accordance with its particular laws. Since the laws of many countries differ in various respects from the patent law of the United States, applicants are advised to seek guidance from specific foreign countries to ensure that patent rights are not lost prematurely.

Applicants also are advised that in the case of inventions made in the United States, the Director of the USPTO must issue a license before applicants can apply for a patent in a foreign country. The filing of a U.S. patent application

GUARDANT HEALTH EX.1009.178

serves as a request for a foreign filing license. The application's filing receipt contains further information and guidance as to the status of applicant's license for foreign filing.

Applicants may wish to consult the USPTO booklet, "General Information Concerning Patents" (specifically, the section entitled "Treaties and Foreign Patents") for more information on timeframes and deadlines for filing foreign patent applications. The guide is available either by contacting the USPTO Contact Center at 800-786-9199, or it can be viewed on the USPTO website at http://www.uspto.gov/web/offices/pac/doc/general/index.html.

For information on preventing theft of your intellectual property (patents, trademarks and copyrights), you may wish to consult the U.S. Government website, http://www.stopfakes.gov. Part of a Department of Commerce initiative, this website includes self-help "toolkits" giving innovators guidance on how to protect intellectual property in specific countries such as China, Korea and Mexico. For questions regarding patent enforcement issues, applicants may call the U.S. Government hotline at 1-866-999-HALT (1-866-999-4258).

LICENSE FOR FOREIGN FILING UNDER

Title 35, United States Code, Section 184

Title 37, Code of Federal Regulations, 5.11 & 5.15

GRANTED

The applicant has been granted a license under 35 U.S.C. 184, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" followed by a date appears on this form. Such licenses are issued in all applications where the conditions for issuance of a license have been met, regardless of whether or not a license may be required as set forth in 37 CFR 5.15. The scope and limitations of this license are set forth in 37 CFR 5.15(a) unless an earlier license has been issued under 37 CFR 5.15(b). The license is subject to revocation upon written notification. The date indicated is the effective date of the license, unless an earlier license of similar scope has been granted under 37 CFR 5.13 or 5.14.

This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

The grant of a license does not in any way lessen the responsibility of a licensee for the security of the subject matter as imposed by any Government contract or the provisions of existing laws relating to espionage and the national security or the export of technical data. Licensees should apprise themselves of current regulations especially with respect to certain countries, of other agencies, particularly the Office of Defense Trade Controls, Department of State (with respect to Arms, Munitions and Implements of War (22 CFR 121-128)); the Bureau of Industry and Security, Department of Commerce (15 CFR parts 730-774); the Office of Foreign AssetsControl, Department of Treasury (31 CFR Parts 500+) and the Department of Energy.

NOT GRANTED

No license under 35 U.S.C. 184 has been granted at this time, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" DOES NOT appear on this form. Applicant may still petition for a license under 37 CFR 5.12, if a license is desired before the expiration of 6 months from the filing date of the application. If 6 months has lapsed from the filing date of this application and the licensee has not received any indication of a secrecy order under 35 U.S.C. 181, the licensee may foreign file the application pursuant to 37 CFR 5.15(b).

GUARDANT HEALTH EX.1009.179

SelectUSA

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The U.S. offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to promote and facilitate business investment. SelectUSA provides information assistance to the international investor community; serves as an ombudsman for existing and potential investors; advocates on behalf of U.S. cities, states, and regions competing for global investment; and counsels U.S. economic development organizations on investment attraction best practices. To learn more about why the United States is the best country in the world to develop technology, manufacture products, deliver services, and grow your business, visit http://www.SelectUSA.gov or call +1-202-482-6800.

GUARDANT HEALTH EX.1009.180

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☒ SMALL ☐ MICRO

APPLICATION AS FILED – PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA	RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*	X $ =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*	X $ =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).			
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))				
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL	

APPLICATION AS AMENDED – PART II

AMENDMENT	03/07/2016	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0	X $40 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	***3	= 0	X $210 =	0
	☐ Application Size Fee (37 CFR 1.16(s))						
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
						TOTAL ADD'L FEE	0

AMENDMENT		(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=	X $ =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=	X $ =	
	☐ Application Size Fee (37 CFR 1.16(s))						
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
						TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.

LIE
WANDA BARNES

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**
If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
U.S. Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

23432 e 2016-07-13

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

Paper No.

Application No.:	**15/063,278**	**Date Mailed:**	**2016-07-13** **PAPER**
First Named Inventor:	James Hicks	Examiner:	LU, FRANK WEI MIN
Attorney Docket No.:	81503-AA/GJG/AWG/AC	Art Unit:	1634
Confirmation No.:	4038	Filing Date:	03/07/2016

Please find attached an Office communication concerning this application or proceeding.

Commissioner for Patents

PTO-90c (Rev.08-06)

Notice of Non-Compliant Amendment (37 CFR 1.121)	Application No. 15/063,278	Applicant(s) HICKS ET AL.

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

The amendment document filed on *07 March, 2016* is considered non-compliant because it has failed to meet the requirements of 37 CFR 1.121 or 1.4. In order for the amendment document to be compliant, correction of the following item(s) is required.

THE FOLLOWING MARKED (X) ITEM(S) CAUSE THE AMENDMENT DOCUMENT TO BE NON-COMPLIANT:

- ☐ 1. Amendments to the specification:
  - ☐ A. Amended paragraph(s) do not include markings.
  - ☐ B. New paragraph(s) should not be underlined.
  - ☐ C. Other _____.
- ☐ 2. Abstract:
  - ☐ A. Not presented on a separate sheet. 37 CFR 1.72.
  - ☐ B. Other _____.
- ☐ 3. Amendments to the drawings:
  - ☐ A. The drawings are not properly identified in the top margin as "Replacement Sheet," "New Sheet," or "Annotated Sheet" as required by 37 CFR 1.121(d).
  - ☐ B. The practice of submitting proposed drawing correction has been eliminated. Replacement drawings showing amended figures, without markings, in compliance with 37 CFR 1.84 are required.
  - ☐ C. Other _____.
- ☒ 4. Amendments to the claims:
  - ☒ A. A complete listing of all of the claims is not present.
  - ☐ B. The listing of claims does not include the text of all pending claims (including withdrawn claims)
  - ☐ C. Each claim has not been provided with the proper status identifier, and as such, the individual status of each claim cannot be identified. Note: the status of every claim must be indicated after its claim number by using one of the following status identifiers: (Original), (Currently amended), (Canceled), (Previously presented), (New), (Not entered), (Withdrawn) and (Withdrawn-currently amended).
  - ☐ D. The claims of this amendment paper have not been presented in ascending numerical order.
  - ☒ E. Other: *Claims 1-71 are not included in the listing of claims*.
- ☐ 5. Other (e.g., the amendment is unsigned or not signed in accordance with 37 CFR 1.4): For further explanation of the amendment format required by 37 CFR 1.121, see MPEP § 714.

TIME PERIODS FOR FILING A REPLY TO THIS NOTICE:

1. Applicant is given **no new time period if the non-compliant amendment is an** after-final amendment or an amendment filed after allowance, or a drawing submission (only) If applicant wishes to resubmit the non-compliant after-final amendment with corrections, the **entire corrected amendment** must be resubmitted.

2. Applicant is given **two months** from the mail date of this notice to supply the correction, if the non-compliant amendment is one of the following: a preliminary amendment, a non-final amendment (including a submission for a request for continued examination (RCE) under 37 CFR 1.114), a supplemental amendment filed within a suspension period under 37 CFR 1.103(a) or (c), and an amendment filed in response to a Quayle action. If any of above boxes 1 to 4 are checked, the correction required is only the corrected section of the non-compliant amendment in compliance with 37 CFR 1.121.

Extensions of time are available under 37 CFR 1.136(a) only if the non-compliant amendment is a non-final amendment or an amendment filed in response to a *Quayle* action.

Failure to timely respond to this notice will result in:

Abandonment of the application if the non-compliant amendment is a non-final amendment or an amendment filed in response to a *Quayle* action; or

Non-entry of the amendment if the non-compliant amendment is a preliminary amendment or supplemental amendment.

Legal Instruments Examiner (LIE), if applicable WANDA BARNES Telephone No: (571)272-7266

GUARDANT HEALTH EX.1009.183

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

PUBLICATION NOTICE

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

OC000000085520335

Title:VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Publication No.US-2016-0251715-A1
Publication Date:09/01/2016

NOTICE OF PUBLICATION OF APPLICATION

The above-identified application will be electronically published as a patent application publication pursuant to 37 CFR 1.211, et seq. The patent application publication number and publication date are set forth above.

The publication may be accessed through the USPTO's publically available Searchable Databases via the Internet at www.uspto.gov. The direct link to access the publication is currently http://www.uspto.gov/patft/.

The publication process established by the Office does not provide for mailing a copy of the publication to applicant. A copy of the publication may be obtained from the Office upon payment of the appropriate fee set forth in 37 CFR 1.19(a)(1). Orders for copies of patent application publications are handled by the USPTO's Office of Public Records. The Office of Public Records can be reached by telephone at (703) 308-9726 or (800) 972-6382, by facsimile at (703) 305-8759, by mail addressed to the United States Patent and Trademark Office, Office of Public Records, Alexandria, VA 22313-1450 or via the Internet.

In addition, information on the status of the application, including the mailing date of Office actions and the dates of receipt of correspondence filed in the Office, may also be accessed via the Internet through the Patent Electronic Business Center at www.uspto.gov using the public side of the Patent Application Information and Retrieval (PAIR) system. The direct link to access this status information is currently http://pair.uspto.gov/. Prior to publication, such status information is confidential and may only be obtained by applicant using the private side of PAIR.

Further assistance in electronically accessing the publication, or about PAIR, is available by calling the Patent Electronic Business Center at 1-866-217-9197.

Office of Data Managment, Application Assistance Unit (571) 272-4000, or (571) 272-4200, or 1-888-786-0101

GUARDANT HEALTH EX.1009.185

Docket No. 81503-AA/GJG/JJC

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	Group Unit : 1634
U.S. Serial No.	:	15/063,278	Examiner: F. Wu
Filed	:	March 7, 2016	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	
Confirmation	:	4038	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
September 13, 2016

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

COMMUNICATION IN RESPONSE TO JULY 13, 2016 NOTICE OF NON-COMPLIANT AMENDMENT (37 C.F.R. 1.121)

This Communication is submitted in response to the July 13, 2016 Notice of Non-Compliant Amendment (37 C.F.R. § 1.121) (hereinafter "July 13, 2016 Notice") issued by the United States Patent and Trademark Office in connection with the above-identified application. A copy of the July 13, 2016 Notice is attached hereto as **Exhibit A.** A response to the July 13, 2016 Notice is due on September 13, 2016. Accordingly, this Communication is being timely filed.

The July 13, 2016 Notice states that the Amendment filed on March 7, 2016 is considered non-compliant because "a complete listing of all the claims is not present." In particular, the July 13, 2016 Notice states that "Claims 1-71 are not included in the listing of claims."

In response, applicants submit the Section titled "Amendments to the Claims" attached hereto as **Exhibit B**, including a complete listing of all of the claims and proper status identifiers in compliance with the requirements of 37 C.F.R. § 1.121.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee is deemed necessary in connection with the filing of this Communication. However, if any additional fee is required, authorization is hereby given to charge the amount of any such additional fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant
Cooper & Dunham LLP (Customer #23432)
30 Rockefeller Plaza, 20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

Joong Yoon Cho 9/13/2016
Date

GUARDANT HEALTH EX.1009.187

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
U.S. Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

23432 e 2016-07-13

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

Paper No.

Application No.:	**15/063,278**	**Date Mailed:**	**2016-07-13** **PAPER**
First Named Inventor:	James Hicks	Examiner:	LU, FRANK WEI MIN
Attorney Docket No.:	81503-AA/GJG/AWG/AC	Art Unit:	1634
Confirmation No.:	4038	Filing Date:	03/07/2016

Please find attached an Office communication concerning this application or proceeding.

Commissioner for Patents

PTO-90c (Rev.08-06)

Applicant: James Hicks et al.
Serial No.: 15/063,278
Filed: March 7, 2016
Exhibit: A

GUARDANT HEALTH EX.1009.188

Notice of Non-Compliant Amendment (37 CFR 1.121)	Application No. 15/063,278	Applicant(s) HICKS ET AL.

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

The amendment document filed on *07 March, 2016* is considered non-compliant because it has failed to meet the requirements of 37 CFR 1.121 or 1.4. In order for the amendment document to be compliant, correction of the following item(s) is required.

THE FOLLOWING MARKED (X) ITEM(S) CAUSE THE AMENDMENT DOCUMENT TO BE NON-COMPLIANT:

☐ 1. Amendments to the specification:
- ☐ A. Amended paragraph(s) do not include markings.
- ☐ B. New paragraph(s) should not be underlined.
- ☐ C. Other _____.

☐ 2. Abstract:
- ☐ A. Not presented on a separate sheet. 37 CFR 1.72.
- ☐ B. Other _____.

☐ 3. Amendments to the drawings:
- ☐ A. The drawings are not properly identified in the top margin as "Replacement Sheet," "New Sheet," or "Annotated Sheet" as required by 37 CFR 1.121(d).
- ☐ B. The practice of submitting proposed drawing correction has been eliminated. Replacement drawings showing amended figures, without markings, in compliance with 37 CFR 1.84 are required.
- ☐ C. Other _____.

☒ 4. Amendments to the claims:
- ☒ A. A complete listing of all of the claims is not present.
- ☐ B. The listing of claims does not include the text of all pending claims (including withdrawn claims)
- ☐ C. Each claim has not been provided with the proper status identifier, and as such, the individual status of each claim cannot be identified. Note: the status of every claim must be indicated after its claim number by using one of the following status identifiers: (Original), (Currently amended), (Canceled), (Previously presented), (New), (Not entered), (Withdrawn) and (Withdrawn-currently amended).
- ☐ D. The claims of this amendment paper have not been presented in ascending numerical order.
- ☒ E. Other: *Claims 1-71 are not included in the listing of claims*.

☐ 5. Other (e.g., the amendment is unsigned or not signed in accordance with 37 CFR 1.4): For further explanation of the amendment format required by 37 CFR 1.121, see MPEP § 714.

TIME PERIODS FOR FILING A REPLY TO THIS NOTICE:

1. Applicant is given **no new time period if the non-compliant amendment is an** after-final amendment or an amendment filed after allowance, or a drawing submission (only) If applicant wishes to resubmit the non-compliant after-final amendment with corrections, the **entire corrected amendment** must be resubmitted.

2. Applicant is given **two months** from the mail date of this notice to supply the correction, if the non-compliant amendment is one of the following: a preliminary amendment, a non-final amendment (including a submission for a request for continued examination (RCE) under 37 CFR 1.114), a supplemental amendment filed within a suspension period under 37 CFR 1.103(a) or (c), and an amendment filed in response to a Quayle action. If any of above boxes 1 to 4 are checked, the correction required is only the corrected section of the non-compliant amendment in compliance with 37 CFR 1.121.

Extensions of time are available under 37 CFR 1.136(a) only if the non-compliant amendment is a non-final amendment or an amendment filed in response to a *Quayle* action.

Failure to timely respond to this notice will result in:

Abandonment of the application if the non-compliant amendment is a non-final amendment or an amendment filed in response to a *Quayle* action; or

Non-entry of the amendment if the non-compliant amendment is a preliminary amendment or supplemental amendment.

Legal Instruments Examiner (LIE), if applicable WANDA BARNES Telephone No: (571)272-7266

GUARDANT HEALTH EX.1009.189

Amendments to the Claims

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

1-71. (Canceled)

72. (New) A method for obtaining genomic copy number information from genomic nucleic acids, comprising:

a) obtaining segments of the genomic nucleic acids;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each tag associated sequence read of step (d); and

f) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome,

GUARDANT HEALTH EX.1009.191

thereby obtaining genomic copy number information from the genomic nucleic acids.

73. (New) The method of claim 72, further comprising estimating a genomic copy number of a region of the genome comprising multiple locations.

74. (New) The method of claim 72, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

75. (New) The method of claim 72, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

76. (New) The method of claim 75, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of

the first region; and

k) obtaining an average of the sum of the counts obtained in step (h) and the counts obtained in step (j).

77. (New) The method of claim 75, wherein the second region of the genome comprises a centromere.

78. (New) The method of claim 72, further comprising l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative genomic copy number of the region of the genome.

79. (New) A method for obtaining mRNA copy number information from mRNA transcripts, comprising:

a) Generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step

(ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information from the mRNA transcripts.

80. (New) The method of claim 72, wherein said tagging the segments comprises:

n) adding a polynucleotide tail to the ends of the segments of the genomic nucleic acids, thereby generating zero-th order

derivative strands comprising the polynucleotide tail;

o) subjecting the zero-th order derivative strands of step (n) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement of the zero-th order derivative strands, thereby generating first order derivative strands;

p) adding a polynucleotide tail to the first order derivative strands, thereby generating a first order derivative strands comprising the polynucleotide tail;

q) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps o) and q) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules.

81. (New) The method of claim 72, wherein said tagging the segments comprises:

r) adding a polynucleotide tail to the ends of the segments of genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail,

s) subjecting the zero-th order derivative strands of step (r) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (r) under conditions that

promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

t) subjecting the product of step (s) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step r) under conditions which promote the formation of only one complement of the product of step (s), wherein the primers of step (s) and the primers of step (t) have different nucleotide sequences, and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (s) and (t) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, thereby generating tagged nucleic acid molecules.

82. (New) The method of claim 81, wherein said adding the polynucleotide tail comprises the use of a terminal transferase.

83. (New) The method of claim 72, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to step (d).

84. (New) The method of claim 72, wherein the tags further comprise a sample tag.

85. (New) The method of claim 84, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the product of step (c).

86. (New) The method of claim 85, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

87. (New) The method of claim 72, wherein the unique tagged nucleic acid molecules are generated from a single cell.

88. (New) The method of claim 83, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR in step (c).

89. (New) The method of claim 88, wherein each of the unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array.

90. (New) The method of claim 83, wherein the amplified unique tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to said sequencing the amplified unique tagged nucleic acid molecules in step (d).

91. (New) The method of claim 90, wherein each of the amplified unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on a microarray.

Electronic Acknowledgement Receipt

EFS ID:	26915733
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Joong Youn Cho
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	13-SEP-2016
Filing Date:	07-MAR-2016
Time Stamp:	19:16:54
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		160913_81503_AA_Communication_JJC.pdf	521309 3d90deb96f426dc054d69ce1c915ca4383787d6f	yes	12

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Miscellaneous Incoming Letter	1	5
	Claims	6	12

Warnings:

The page size in the PDF is too large. The pages should be 8.5 x 11 or A4. If this PDF is submitted, the pages will be resized upon entry into the Image File Wrapper and may affect subsequent processing

Information:

Total Files Size (in bytes):	521309

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☒ SMALL ☐ MICRO

APPLICATION AS FILED – PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA	RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*	X $ =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*	X $ =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).			
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))				
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL	

APPLICATION AS AMENDED – PART II

AMENDMENT 09/13/2016	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0	X $40 =	0
Independent (37 CFR 1.16(h))	* 2	Minus	***3	= 0	X $210 =	0
☐ Application Size Fee (37 CFR 1.16(s))						
☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
					TOTAL ADD'L FEE	0

AMENDMENT	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
Total (37 CFR 1.16(i))	*	Minus	**	=	X $ =	
Independent (37 CFR 1.16(h))	*	Minus	***	=	X $ =	
☐ Application Size Fee (37 CFR 1.16(s))						
☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
					TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.

LDRC
ADRIANE WINSTON

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**
If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

No fee is deemed necessary in connection with the filing of this Preliminary Amendment. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

Abbie Chung 6/19/2017
Abbie Chung Date

REMARKS

By this Preliminary Amendment, applicants have amended the government support clause in the specification to comply with 37 C.F.R. §401.14(f)(4) and 35 U.S.C. §202(c)(6).

Applicants maintain that the amendments presented hereinabove raise no issue of new matter and respectfully request that this Preliminary Amendment be entered.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

Amendments to the Specification

Please amend the paragraph on page 1, lines 8-11 of the specification as follows:

~~The~~This invention ~~disclosed herein~~ was ~~supported by the~~made with government support under grant ~~No.~~ W81XWH-09-1-0591 ~~from the Department of Defense~~awarded by Army Medical Research Materiel Command. ~~Accordingly, the U.S. Government~~The government has certain rights ~~to this~~in the invention.

Docket No. 81503-AA/GJG/AC

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
Serial No.	:	15/063,278	Examiner: Frank Lu
Filed	:	March 7, 2016	Group Art Unit: 1634
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	
Conf. No.	:	4038	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
June 19, 2017

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

FOURTH PRELIMINARY AMENDMENT

This Preliminary Amendment is submitted in connection with the above-identified application prior to the mailing of a first Office Action.

Amendments to the Specification begin on page 2 of this paper.

Remarks begin on page 3 of this paper.

Electronic Acknowledgement Receipt

EFS ID:	29537065
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	19-JUN-2017
Filing Date:	07-MAR-2016
Time Stamp:	14:48:23
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		170619_81503-AA_Preliminary_Amendment_AC.pdf	171623 d4de6a76cf0b5c133a1fd1d0cc4cbeb6dcce15b2	yes	4

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Preliminary Amendment	4	4
	Applicant Arguments/Remarks Made in an Amendment	3	3
	Specification	2	2
	Preliminary Amendment	1	1
Warnings:			
Information:			
	Total Files Size (in bytes):	171623	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	5,639,603	06-17-1997	Dower et al.
	2	7,537,897	05-26-2009	Brenner et al.
	3	7,622,281	11-24-2009	Ronaghi et al.
	4	8,663,917	03-04-2014	Wigler et al.
	5	8,694,263	04-08-2014	Wigler et al.
	6	6,569,617	05-27-2013	Wigler et al.
	7	7,531,307	05-12-2009	Wigler et al.
	8	8,173,871	09-25-2012	Hannon et al.
	9	8,554,488	10-08-2013	Wigler et al.
	10	7,700,324	04-20-2010	Issa et al.
	11	6,172,214	01-09-2001	Brenner et al.
	12	2004/110153	06-10-2004	Dong et al.
	13	2007/172873	07-26-2007	Brenner et al.
	14	2010/178655	07-15-2010	Hamilton et al.
	15	2006/0073506	04-06-2006	Christians et al.
	16	2005/0032095	02-10-2005	Wigler et al.
	17	2005/0266444	12-01-2005	Wigler et al.
	18	2007/0207481	09-06-2007	Wigler et al.
	19	2010/0227768	09-10-2010	Wigler et al.
	20	2012/0149593	06-14-2012	Hicks et al.
	21	2012/0328607	12-27-2012	Hicks et al.
	22	2013/0179999	07-11-2013	Hannon et al.

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
First IDS Exhibit A

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	23	2014/0025307	01-23-2014	Wigler et al.
	24	2006/0035258	02-16-2006	Tadakamalla et al.
	25	2009/0137402	05-28-2009	Wang et al.
	26	2010/0203538	08-12-2010	Dube et al.

FOREIGN PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Foreign Patent Document Country Code[3] Number[4] Kind Code[5] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	T[6]
	27	WO 2007/062445	06-07-2007	Alexander Seitz	
	28	WO 2012/054873	04-26-2012	Hicks et al.	

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	29	February 13, 2014 Extended European Search Report, issued in connection with European Patent Application No. 11835243.4	
	30	April 23, 2013 International Preliminary Report on Patentability issued in connection with PCT International Application No. PCT/US2011/057350 (WO 2012/054873)	
	31	June 26, 2012 International Search Report issued in connection wth PCT International Application No. PCT/US2011/057350 (WO 2012/054873)	
	32	Miner et al., "Molecular barcodes detect redundancy and contamination in hairpin-bisulfite PCR," Nucleic Acids Res. vol. 32 no. 17 (2004), p. 1-4	
	33	Casbon et al., "A method for counting PCT template molecules with application to next-generation sequencing", Nucleic Acids Res. vol. 39 no. 12 (July 2011)	
	34	Cheung V.G. & Nelson S.F., "Whole genome amplification using a degenerate oligonucleotide primer allows hundreds of genotypes to be performed on less than one nanogram of genomic DNA," PNAS vol. 93 no. 25 (Dec. 1996), p. 14676–79	
	35	Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," Science vol. 323 no. 5910 (Jan. 2009), p. 133-38	
	36	Supporting Online Material for Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," accessible at www.sciencemag.org/cgi/content/full/1162986/DC1, (published Nov. 20, 2008)	
	37	Fu G.K. et al., "Counting individual DNA molecules by the stochastic attachment of diverse labels," PNAS vol. 108 no. 22 (May 2011), p. 9026-31	
	38	McCloskey M.L. et al, "Encoding PCR Products with Batch-stamps and Barcodes," Biochem Genet. vol. 45 (Dec. 2007), p. 761-67	
	39	Parameswaran et al., "A pyrosequencing-tailored nucleotide barcode design unveils opportunities for large-scale sample multiplexing," Nucleic Acids Res. vol. 35 no. 19 (2007)	
	40	Paunio T. et al., "Preimplantation diagnosis by whole genome amplification, PCT amplification, and solid-phase minisequencing of blastomere DNA," Clinical Chem. vol. 42 no. 9 (1996), p. 1382-90	
	41	Technology Backgrounder: Single Molecule Real Time (SMRT™) DNA Sequencing, Pacific Bioscience (2009)	
	42	Islam et al., "Characterization of the single-cell transcriptional landscape by highly multiplex RNA-seq," Genome Res. vol. 21 no. 7 (July 2011) p. 1160-67	

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	43	Kivioja et al., "Counting absolute numbers of molecules using unique molecular identifiers, Nature Methods vol. 9 (2012), p. 72-74	
	44	Shiroguchi at al., "Digital RNA sequencing minimizes sequence-dependent bias and amplification noise with optimized single-molecule barcodes," PNAS vol. 109 no. 4 (2012), p. 1347-52	
	45	Castle et al., "DNA copy number, including telomeres and mitochondria, assayed using next-generation sequencing," BMC Genomics vol. 11 no. 244 (2010)	
	46	Craig et al., "Identification of Genetic Variants Using Bar-coded Multiplexed Sequencing," Nature Methods vol. 5 no. 10 (Oct. 2008), p. 887-93.	

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

Dkt. 81503-AA/GJG/DH

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants:	James Hicks et al.	
Serial No.:	15/063,278	Examiner: Frank Wei Min Lu
Filed :	March 7, 2016	Art Unit: 1634
Conf. No. :	4038	
For :	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
New York, New York 10112
September 6, 2017

By EFS Web
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

SECOND INFORMATION DISCLOSURE STATEMENT

In accordance with the duty of disclosure under 37 C.F.R. § 1.56, applicant directs the Examiner's attention to the following items listed on the Form PTO-1449 (Substitute) attached hereto as **Second IDS, Exhibit A.**

This Information Disclosure Statement is being submitted pursuant to 37 C.F.R. §1.97(b)(3) before the mailing of a first office action on the merits and no fee is deemed necessary with the filing of this Information Disclosure Statement.

Reference items 1-5 are U.S. Patents and U.S. patent application publications. Pursuant to 37 C.F.R. §1.98(a)(2), copies of items 1-5 are not being submitted.

1. U.S. Patent No. 9,404,156 B2, issued August 2, 2016 (Hicks et al.);

2. U.S. Patent No. 9,476,095 B2, issued October 25, 2016 (Vogelstein et al.);

3. U.S. Patent No. 9,487,829 B2, issued November 8, 2016 (Vogelstein et al.);

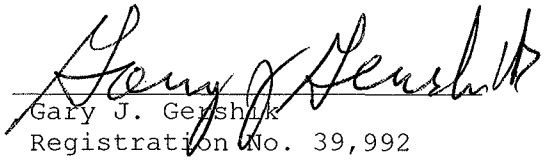
4. U.S. Patent Application Publication No. 2016/0215333 A1, published July 28, 2016 (Vogelstein et al.); and

5. U.S. Patent Application Publication No. 2017/0051347 A1, published February 23, 2017 (Vogelstein et al.).

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the telephone number provided below.

No fee is deemed necessary in connection with the filing of this Information Disclosure Statement. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorneys for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on September 6, 2017.



Darren Haber

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
INFORMATION DISCLOSURE STATEMENT (Use several sheets if necessary)	**Application Number**	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	9,404,156 B2	08-02-2016	Hicks et al.
	2	9,476,095 B2	10-25-2016	Vogelstein et al.
	3	9,487,829 B2	11-08-2016	Vogelstein et al.
	4	2016/0215333 A1	07-28-2016	Vogelstein et al.
	5	2017/0051347 A1	02-23-2017	Vogelstein et al.

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
Second IDS Exhibit A

Electronic Acknowledgement Receipt

EFS ID:	30288336
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	06-SEP-2017
Filing Date:	07-MAR-2016
Time Stamp:	17:52:37
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		170906_81503-AA_FirstIDS_DH.pdf	489943 a33bae9e75fa14686b82ff95ec5406fcb4547924	yes	10

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Transmittal Letter	1	6
	Information Disclosure Statement (IDS) Form (SB08)	7	10

Warnings:					
Information:					
2		170906_81503-AA_SecondIDS_DH.pdf	121295 6561640e26a58441697dc6901c3ca9737fdb2c8d	yes	4

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Transmittal Letter	1	3
	Information Disclosure Statement (IDS) Form (SB08)	4	4

Warnings:	
Information:	
Total Files Size (in bytes):	611238

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

Dkt. 81503-AA/GJG/DH

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants: James Hicks et al.

Serial No.: 15/063,278 Examiner: Frank Wei Min Lu

Filed : March 7, 2016 Art Unit: 1634

Conf. No. : 4038

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
New York, New York 10112
September 6, 2017

By EFS Web
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

FIRST INFORMATION DISCLOSURE STATEMENT

In accordance with the duty of disclosure under 37 C.F.R. § 1.56, applicant directs the Examiner's attention to the following items listed on the Form PTO-1449 (Substitute) attached hereto as **First IDS, Exhibit A.**

This Information Disclosure Statement is being submitted pursuant to 37 C.F.R. §1.97(b)(3) before the mailing of a first office action on the merits and no fee is deemed necessary with the filing of this Information Disclosure Statement.

Reference items 1-26 are U.S. Patents and U.S. patent application publications. Pursuant to 37 C.F.R. §1.98(a)(2), copies of items 1-26 are not being submitted.

The above identified application claims benefit under 35 U.S.C. §120 of U.S. Serial No. 13/278,333, filed October 21, 2011. Copies of references 27-46 listed below have previously been submitted to, or cited by the U.S. Patent Office in connection with U.S. Serial No. 13/278,333. Therefore, in accordance with 37 C.F.R. §1.98(d), copies of the previously submitted or cited references are not provided.

1. U.S. Patent No. 5,639,603, issued June 17, 1997 (Dower et al.);

2. U.S. Patent No. 7,537,897, issued May 26, 2009 (Brenner et al.);

3. U.S. Patent No. 7,622,281, issued November 24, 2009 (Ronaghi et al.);

4. U.S. Patent No. 8,663,917, issued November 24, 2009 (Wigler et al.);

5. U.S. Patent No. 8,694,263, issued April 8, 2014 (Wigler et al.);

6. U.S. Patent No. 6,569,617, issued May 27, 2013 (Wigler et al.);

7. U.S. Patent No. 7,531,307, issued May 12, 2009 (Wigler et al.);

8. U.S. Patent No. 8,173,871, issued September 25, 2012 (Hannon et al.);

9. U.S. Patent No. 8,554,488, issued October 8, 2013 (Wigler et al.);

10. U.S. Patent No. 7,700,324, issued April 20, 2010 (Issa et al.);

11. U.S. Patent No. 6,172,214, January 9, 2001 (Brenner et al.);

12. U.S. Patent Application Publication No. 2004/110153, published June 10, 2004 (Dong et al.);

13. U.S. Patent Application Publication No. 2007/172873, published July 26, 2007 (Brenner et al.);

14. U.S. Patent Application Publication No. 2010/178655, published July 15, 2010 (Hamilton et al.);

15. U.S. Patent Application Publication No. 2006/0073506, April 6, 2006 (Christians et al.);

16. U.S. Patent Application Publication No. 2005/0032095, published February 10, 2005 (Wigler et al.);

17. U.S. Patent Application Publication No. 2005/0266444, published December 1, 2005 (Wigler et al.);

18. U.S. Patent Application Publication No. 2007/0207481, published September 6, 2007 (Wigler et al.);

19. U.S. Patent Application Publication No. 2010/0227768, published September 10, 2010 (Wigler et al.);

20. U.S. Patent Application Publication No. 2012/0149593, published June 14, 2012 (Hicks et al.);

21. U.S. Patent Application Publication No. 2012/0328607, published December 27, 2012 (Hicks et al.);

22. U.S. Patent Application Publication No. 2013/0179999, published July 11, 2013 (Hannon et al.);

23. U.S. Patent Application Publication No. 2014/0025307, published January 23, 2014 (Wigler et al.);

24. U.S. Patent Application Publication No. 2006/0035258, published February 16, 2006 (Tadakamalla et al.);

25. U.S. Patent Application Publication No. 2009/0137402, published May 28, 2009 (Wang et al.);

26. U.S. Patent Application Publication No. 2010/0203538, published

August 12, 2010 (Dube et al.);

27. PCT International Publication No. WO 2007/062445, published June 7, 2007 to Alexander Seitz;

28. PCT International Publication No. WO 2012/054873, published April 26 2012 to Hicks et al.;

29. February 13, 2014 Extended European Search Report, issued in connection with European Patent Application No. 11835243.4;

30. April 23, 2013 International Preliminary Report on Patentability issued in connection with PCT International Application No. PCT/US2011/057350 (WO 2012/054873);

31. June 26, 2012 International Search Report issued in connection with PCT International Application No. PCT/US2011/057350 (WO 2012/054873);

32. Miner et al., "Molecular barcodes detect redundancy and contamination in hairpin-bisulfite PCR," Nucleic Acids Res. vol. 32 no. 17 (2004), p. 1-4;

33. Casbon et al., "A method for counting PCT template molecules with application to next-generation sequencing", Nucleic Acids Res. vol. 39 no. 12 (July 2011);

34. Cheung V.G. & Nelson S.F., "Whole genome amplification using a degenerate oligonucleotide primer allows hundreds of genotypes to be performed on less than one nanogram of genomic DNA," PNAS vol. 93 no. 25 (Dec. 1996), p. 14676-79;

35. Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," Science vol. 323 no. 5910 (Jan. 2009), p. 133-38;

36. Supporting Online Material for Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," accessible at www.sciencemag.org/cgi/content/full/1162986/DC1, (published Nov. 20, 2008);

37. Fu G.K. et al., "Counting individual DNA molecules by the stochastic attachment of diverse labels," PNAS vol. 108 no. 22 (May 2011), p. 9026-31;

38. McCloskey M.L. et al, "Encoding PCR Products with Batch-stamps and Barcodes," Biochem. Genet. vol. 45 (Dec. 2007), p. 761-67;

39. Parameswaran et al., "A pyrosequencing-tailored nucleotide barcode design unveils opportunities for large-scale sample multiplexing," Nucleic Acids Res. vol. 35 no. 19 (2007);

40. Paunio T. et al., "Preimplantation diagnosis by whole genome amplification, PCT amplification, and solid-phase minisequencing of blastomere DNA," Clinical Chem. vol. 42 no. 9 (1996), p. 1382-90;

41. Technology Backgrounder: Single Molecule Real Time (SMRTTM) DNA Sequencing, Pacific Bioscience (2009);

42. Islam et al., "Characterization of the single-cell transcriptional landscape by highly multiplex RNA-seq," Genome Res. vol. 21 no. 7 (July 2011) p. 1160-67;

43. Kivioja et al., "Counting absolute numbers of molecules using unique molecular identifiers, Nature Methods vol. 9 (2012), p. 72-74;

44. Shiroguchi at al., "Digital RNA sequencing minimizes sequence-dependent bias and amplification noise with optimized single-molecule barcodes," PNAS vol. 109 no. 4 (2012), p. 1347-52;

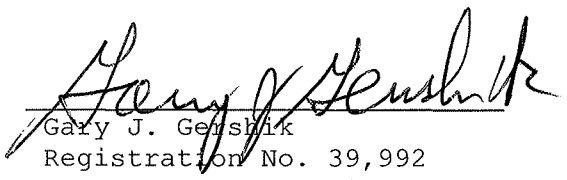
45. Castle et al., "DNA copy number, including telomeres and mitochondria, assayed using next-generation sequencing," BMC Genomics vol. 11 no. 244 (2010); and

46. Craig et al., "Identification of Genetic Variants Using Bar-coded Multiplexed Sequencing," Nature Methods vol. 5 no. 10 (Oct. 2008), p. 887-93.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the telephone number provided below.

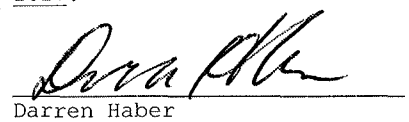
No fee is deemed necessary in connection with the filing of this Information Disclosure Statement. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorneys for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on September 6, 2017.



Darren Haber

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 01/18/2018

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
01/18/2018	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.223

Office Action Summary	Application No. 15/063,278	Applicant(s) HICKS ET AL.	
	Examiner FRANK LU	Art Unit 1634	AIA (First Inventor to File) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 2 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *3/7/2016*.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5)☒ Claim(s) *68-87* is/are pending in the application.

5a) Of the above claim(s) _____ is/are withdrawn from consideration.

6)☐ Claim(s) _____ is/are allowed.

7)☐ Claim(s) _____ is/are rejected.

8)☐ Claim(s) _____ is/are objected to.

9)☒ Claim(s) *68-87* are subject to restriction and/or election requirement.

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to PPHfeedback@uspto.gov.

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)

2) ☐ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date _____.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .

4) ☐ Other: _____.

GUARDANT HEALTH EX.1009.224

DETAILED ACTION

Preliminary Amendment

1. The preliminary amendment filed on March 7, 2016 has been entered. Since claims 1-67 have been canceled and there are no claims 68-71 in this instant application, newly added claims 72-91 in the preliminary amendment should be renumbered as claims 68-87.

Election/Restrictions

2. This application contains claims directed to the following patentably distinct species:

(1) subjecting the zero-th order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement of the zero-th order derivative strands (claim 76)

(2) subjecting the zero-th order derivative strands to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the ligation of a primer to the 5' ends of the zero-th order derivative strands (claim 77)

The species are independent or distinct because these species are directed to different reactions of the zero-th order derivative strands which have different properties. In addition, these species are not obvious variants of each other based on the current record.

Applicant is required under 35 U.S.C. 121 to elect a single disclosed species, or a single grouping of patentably indistinct species, for prosecution on the merits to which the claims shall

be restricted if no generic claim is finally held to be allowable. Currently, generic claims are claims 68-75 and 78-87.

There is a search and/or examination burden for the patentably distinct species as set forth above because the searches for these species are not overlap.

Applicant is advised that the reply to this requirement to be complete must include (i) an election of a species to be examined even though the requirement may be traversed (37 CFR 1.143) **and (ii) identification of the claims encompassing the elected species or grouping of patentably indistinct species**, including any claims subsequently added. An argument that a claim is allowable or that all claims are generic is considered nonresponsive unless accompanied by an election.

The election may be made with or without traverse. To preserve a right to petition, the election must be made with traverse. If the reply does not distinctly and specifically point out supposed errors in the election of species requirement, the election shall be treated as an election without traverse. Traversal must be presented at the time of election in order to be considered timely. Failure to timely traverse the requirement will result in the loss of right to petition under 37 CFR 1.144. If claims are added after the election, applicant must indicate which of these claims are readable on the elected species or grouping of patentably indistinct species.

Should applicant traverse on the ground that the species, or groupings of patentably indistinct species from which election is required, are not patentably distinct, applicant should submit evidence or identify such evidence now of record showing them to be obvious variants or clearly admit on the record that this is the case. In either instance, if the examiner finds one of

the species unpatentable over the prior art, the evidence or admission may be used in a rejection under 35 U.S.C. 103 or pre-AIA 35 U.S.C. 103(a) of the other species.

Upon the allowance of a generic claim, applicant will be entitled to consideration of claims to additional species which depend from or otherwise require all the limitations of an allowable generic claim as provided by 37 CFR 1.141.

Applicant is reminded that upon the cancellation of claims to a non-elected invention, the inventorship must be corrected in compliance with 37 CFR 1.48(a) if one or more of the currently named inventors is no longer an inventor of at least one claim remaining in the application. A request to correct inventorship under 37 CFR 1.48(a) must be accompanied by an application data sheet in accordance with 37 CFR 1.76 that identifies each inventor by his or her legal name and by the processing fee required under 37 CFR 1.17(i).

3. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993)(See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Frank W Lu /
Primary Examiner, Art Unit 1634
January 9, 2018

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

BIB DATA SHEET

CONFIRMATION NO. 4038

SERIAL NUMBER	FILING or 371(c) DATE	CLASS	GROUP ART UNIT	ATTORNEY DOCKET NO.
15/063,278	03/07/2016 RULE	435	1634	81503-AA/GJG/AWG/AC

APPLICANTS

INVENTORS

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

**** CONTINUING DATA ***************************

This application is a CON of 13/278,333 10/21/2011 PAT 9404156
which claims benefit of 61/510,579 07/22/2011
and claims benefit of 61/406,067 10/22/2010

**** FOREIGN APPLICATIONS ***************************

**** IF REQUIRED, FOREIGN FILING LICENSE GRANTED **** ** SMALL ENTITY **
03/22/2016

Foreign Priority claimed ☐ Yes ☑ No 35 USC 119(a-d) conditions met ☐ Yes ☐ No Verified and Acknowledged /FRANK WEI MIN LU/ Examiner's Signature	☐ Met after Allowance Initials	STATE OR COUNTRY	SHEETS DRAWINGS	TOTAL CLAIMS	INDEPENDENT CLAIMS
		NY	11	20	2

ADDRESS

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112
UNITED STATES

TITLE

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

FILING FEE RECEIVED		
730	FEES: Authority has been given in Paper No._____ to charge/credit DEPOSIT ACCOUNT No._____ for following:	☐ All Fees ☐ 1.16 Fees (Filing) ☐ 1.17 Fees (Processing Ext. of time) ☐ 1.18 Fees (Issue) ☐ Other _____ ☐ Credit

Docket No. 5981/81503-AA/GJG/ADR/AMM

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
March 16, 2018

VIA EFS-WEB
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

RESPONSE TO JANUARY 18, 2018 OFFICE ACTION

This Response is being submitted in response to the January 18, 2018 Office Action issued by the United States Patent and Trademark Office in connection with the above-identified application. A response to the January 18, 2018 Office Action is due March 18, 2018. Accordingly, this Response is being timely filed.

Remarks begin on page 2 of this paper.

REMARKS

Claims 68-87 are pending in the subject application. Applicants have not herein amended, added, or canceled any claim. Accordingly, claims 68-87 are pending in the subject application.

Election of Species

In the January 18, 2018 Office Action, the Examiner required applicants to elect a single species from among the species listed below for initiating prosecution on the merits and to which the claims shall be restricted if no generic claim is finally held to be allowable:

I. subjecting the zero-th order derivative strands to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement, thereby generating first order derivative strands (renumbered claim 76);

II. subjecting the zero-th order derivative strands of step (a) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands (renumbered claim 77).

The Examiner further indicated that claims 68-75 and 78-87 are generic.

Applicants' Response

In response, applicants elect for initiating examination, **species II**, *i.e.* subjecting the zero-th order derivative strands of step (a) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands (renumbered claim 77).

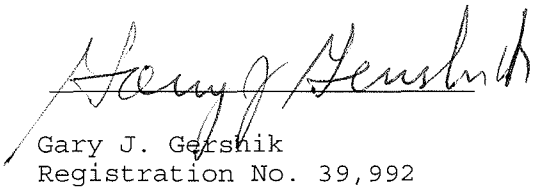
Claims 68-75 and 77-87 encompass the elected species.

Applicants understand that once a generic claim is allowable, the Examiner will examine non-elected species that require all of the limitations of the generic claim.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

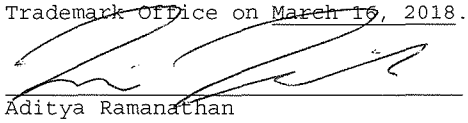
No fee is deemed necessary in connection with the filing of this Response. If any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on March 16, 2018.



Aditya Ramanathan

Docket No. 5981/81503-AA/GJG/ADR/AMM

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
March 16, 2018

VIA EFS-WEB
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

THIRD INFORMATION DISCLOSURE STATEMENT

In accordance with their duty of disclosure under 37 C.F.R. § 1.56, applicant directs the Examiner's attention to the following documents which are listed on Form PTO-1449 **(Exhibit A)** and are also listed below.

This Information Disclosure Statement is being submitted before the mailing of the first Office Action on the merits. Thus, no fee is required and this Information Disclosure Statement should be entered and considered.

Reference item **1** is a U.S. Patent. Pursuant to 37 C.F.R 1.98(a)(2)(ii), a copy of reference **1** is not being submitted.

Each of reference items **2-11** were issued in connection with the examination of foreign counterpart applications to the instant application.

The above identified application claims benefit under 35 U.S.C. §120 of U.S. Serial No. 13/278,333, filed October 21, 2011. Copies of references **10** and **11** listed below have previously been submitted to, or cited by the U.S. Patent Office in connection with U.S. Serial No. 13/278,333. Therefore, in accordance with 37 C.F.R. §1.98(d), copies of the previously submitted or cited references are not provided.

Copies of reference items **2-9** are attached hereto as **Exhibits 1-8,** respectively.

1. U.S. Patent No. 6,013,445, issued January 11, 2000 (Albrecht et al.);

2. October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4 **(Exhibit 1)**;

3. February 16, 2018 Amendment under Rule 71(3) EPC to October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4 **(Exhibit 2)**;

4. December 8, 2017 Office Action issued by the Canadian Intellectual Property Office in connection with Canadian Patent Application 2,815,076 **(Exhibit 3)**;

5. May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807 **(Exhibit 4)**;

6. April 19, 2017 Response to May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807 **(Exhibit 5)**;

7. April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807 **(Exhibit 6)**;

8. May 2, 2017 Response to April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807 **(Exhibit 7)**;

9. September 7, 2017 Notice of Grant for Patent issued by IP Australia in connection with Australian Patent Application 2011316807 **(Exhibit 8)**;

10. November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4; and

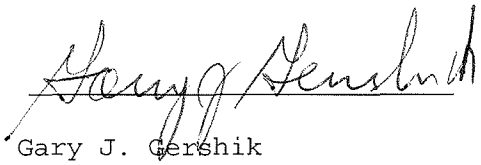
11. June 27, 2016 Amendment under Rule 94(3) EPC to November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4.

The Examiner is respectfully requested to make of record each item listed on the substitute Form PTO-1449 by initialing and dating the attached substitute Form PTO-1449, and returning a copy of the initialed and dated form to applicants' undersigned attorney.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

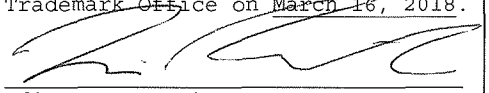
No fee is deemed necessary in connection with the filing of this Third Information Disclosure Statement. If any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark ~~Office~~ on ~~March 16~~, 2018.



Aditya Ramanathan

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
INFORMATION DISCLOSURE STATEMENT (Use several sheets if necessary)	**Application Number**	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/ADR/AMM

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	6,013,445	01-11-2000	Albrecht et al.

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	2	October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	3	February 16, 2018 Amendment under Rule 71(3) EPC to October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	4	December 8, 2017 Office Action issued by the Canadian Intellectual Property Office in connection with Canadian Patent Application 2,815,076	
	5	May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807	
	6	April 19, 2017 Response to May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807	
	7	April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807	
	8	May 2, 2017 Response to April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807	
	9	September 7, 2017 Notice of Grant for Patent issued by IP Australia in connection with Australian Patent Application 2011316807	
	10	November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	11	June 27, 2016 Amendment under Rule 94(3) EPC to November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	

EXAMINER SIGNATURE	DATE CONSIDERED

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
Third IDS Exhibit A

Electronic Acknowledgement Receipt

EFS ID:	32082346
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	16-MAR-2018
Filing Date:	07-MAR-2016
Time Stamp:	19:22:25
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		180316_81503_AA_Response_AMM.pdf	108016 33c19912ccf3659ebf12affd8550bc91b9d39afd	yes	4

	Multipart Description/PDF files in .zip description			
	Document Description	**Start**	**End**	
	Response to Election / Restriction Filed	1	1	
	Applicant Arguments/Remarks Made in an Amendment	2	4	

Warnings:					
Information:					
2	Transmittal Letter	180316_81503_AA_Third_IDS_Transmittal_AMM.pdf	131014 3da8d1f0fca8555a2719507ce24b09e73169b8b3	no	4
Warnings:					
Information:					
3	Information Disclosure Statement (IDS) Form (SB08)	180316_81503_AA_1449_AMM.pdf	87087 05b188b7c5eb4064d6581b84e93c118fda0dac83	no	1
Warnings:					
Information:					
This is not an USPTO supplied IDS fillable form					
4	Non Patent Literature	180316_81503_AA_Exhibit1_AMM.pdf	1322928 1335c4e4d0eed1fb021aa89580251d33c4d7ce4c	no	24
Warnings:					
Information:					
5	Non Patent Literature	180316_81503_AA_Exhibit2_AMM.pdf	6974055 039b85c811961cb615fc55af45ed2d6f7ec9e6bd	no	148
Warnings:					
Information:					
6	Non Patent Literature	180316_81503_AA_Exhibit3_AMM.pdf	510153 b540163eb4deb502ca95cc5185ac7a45425bb2d9	no	6
Warnings:					
Information:					
7	Non Patent Literature	180316_81503_AA_Exhibit4_AMM.pdf	448027 850fb6f9dfc57dbede5bc125123c217576b28e94	no	3

Warnings:					
Information:					
8	Non Patent Literature	180316_81503_AA_Exhibit5_AMM.pdf	1922296 0f56eccc19c26345e8353daeeb69da95f7400194	no	38
Warnings:					
Information:					
9	Non Patent Literature	180316_81503_AA_Exhibit6_AMM.pdf	243406 bf31467a23da24dc055c78a0e9304e502253f481	no	4
Warnings:					
Information:					
10	Non Patent Literature	180316_81503_AA_Exhibit7_AMM.pdf	1711133 a80f6c9c7ea90556a0a48885e186d9a8ed909c82	no	38
Warnings:					
Information:					
11	Non Patent Literature	180316_81503_AA_Exhibit8_AMM.pdf	1437978 6fc93e7cd329adaa76cb9a9344349dbe4984749	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	14896093		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 04/17/2018

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
04/17/2018	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.242

Office Action Summary	Application No. 15/063,278	Applicant(s) HICKS ET AL.	
	Examiner FRANK LU	Art Unit 1634	AIA (First Inventor to File) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *3/16/2018*.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5)☒ Claim(s) *68-87* is/are pending in the application.

5a) Of the above claim(s) *76* is/are withdrawn from consideration.

6)☐ Claim(s) _____ is/are allowed.

7)☒ Claim(s) *68-75 and 77-87* is/are rejected.

8)☐ Claim(s) _____ is/are objected to.

9)☐ Claim(s) _____ are subject to restriction and/or election requirement.

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to PPHfeedback@uspto.gov.

Application Papers

10)☒ The specification is objected to by the Examiner.

11)☒ The drawing(s) filed on *3/7/2016* is/are: a)☐ accepted or b)☒ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)

2) ☒ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date *9/6/2017 and 3/16/2018*.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .

4) ☐ Other: _____.

GUARDANT HEALTH EX.1009.243

DETAILED ACTION

Election/Restrictions

1. Applicant's election of species (2) (subjecting the zero-th order derivative strands to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the ligation of a primer to the 5' ends of the zero-th order derivative strands, see claim 77) in the reply filed on March 16, 2018 is acknowledged. Because applicant did not distinctly and specifically point out the supposed errors in the restriction requirement, the election has been treated as an election without traverse (MPEP § 818.01(a)). Claims 68-75 and 77-87 will be examined.

Drawings

2. Some letters in Figures 7, 9, and 10 are difficult to be recognized. Applicant is required to furnish a drawing under 37 CFR 1.81(c). No new matter may be introduced in the required drawing. Each drawing sheet submitted after the filing date of an application must be labeled in the top margin as either "Replacement Sheet" or "New Sheet" pursuant to 37 CFR 1.121(d).

Specification

3. The disclosure is objected to because of the following informalities: (1) since case 13/273,333 now is US Patent No. 9,404,156 B2, applicant is required to update this information in the first paragraph of the specification; (2) there are Figures A, B, and C. However, BRIEF DESCRIPTION OF THE FIGURES only describes Figure 5; and (3) there are some nucleotide sequences having more than 10 nucleotides in Figures 8 and 9. However, there are no SEQ ID

NOs for these nucleotide sequences in Figures 8 and 9 or BRIEF DESCRIPTION OF THE FIGURES related to Figures 8 and 9 do not describe these nucleotide sequences.

Appropriate correction is required.

Claim Objections

4. Claim 68 is objected to because of the following informality: "each tag associated sequence read of step (d)" in step e) should be "each of the tag associated sequence reads in step (d)".

5. Claim 81 is objected to because of the following informality: "said sequencing of the product of step (c)" should be "said sequencing of the amplified tagged nucleic acid molecules of step (c)".

6. Claim 85 or 89 is objected to because of the following informality: "subject to" should be "subjected to".

Appropriate correction is required.

Claim Rejections - 35 USC § 112

7. The following is a quotation of 35 U.S.C. 112(b):

> (b) CONCLUSION.—The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the inventor or a joint inventor regards as the invention.

The following is a quotation of 35 U.S.C. 112 (pre-AIA), second paragraph:
> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

8. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 112(b) or 35 U.S.C. 112 (pre-AIA), second paragraph, as being indefinite for failing to particularly point out and distinctly claim the

subject matter which the inventor or a joint inventor, or for pre-AIA the applicant regards as the invention.

9. Claim 68 is rejected as vague and indefinite in view of step b) because it is unclear what means "corresponding to". Does the phrase "corresponding to" mean "comprising" or mean something else? Furthermore, it is unclear how genomic copy number information can obtained from the genomic nucleic acids based on step f). Please clarify.

10. Claim 69 recites the limitation "the genome comprising multiple locations" in the claim. There is insufficient antecedent basis for this limitation in the claim because there is no phrase "a genome comprising multiple locations" in claim 68. Please clarify.

11. Claim 70 or 71 or 72 or 74 recites the limitation "the counts obtained in step (f)" in the claim. There is insufficient antecedent basis for this limitation in the claim because there is no word "counts" in step f) of claim 68. Please clarify.

12. Claim 75 is rejected as vague and indefinite in view of step b) because it is unclear how mRNA copy number information can obtained from the mRNA transcripts based on step e). Please clarify.

13. Claim 77 is rejected as vague and indefinite because it is unclear what is the relationship between tagged nucleic acid molecules in the end of the claim and unique tagged nucleic acid molecules in step b) of claim 68. Please clarify.

Claim Rejections - 35 USC § 101

14. 35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

15. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 101 because

The claimed invention is directed to a judicial exception (i.e., a law of nature, a natural phenomenon, or an abstract idea) without significantly more. Claims 68-74 and 77-87 are directed to a process of obtaining genomic copy number information from genomic nucleic acids while claim 75 is directed to a process of obtaining mRNA copy number information from mRNA transcripts. The claims do not include additional elements that are sufficient to amount to significantly more than the judicial exception because:

The Supreme Court has identified a number of considerations for determining whether a claim with additional elements amounts to significantly more than the judicial exception itself. The following are examples of these considerations, which are not intended to be exclusive or limiting. Limitations that may be enough to qualify as "significantly more" when recited in a claim with a judicial exception include:

• Applying the judicial exception with, or by use of, a particular machine;

• Effecting a transformation or reduction of a particular article to a different state or thing;

• Adding a specific limitation other than what is well-understood, routine and conventional in the field, or adding unconventional steps that confine the claim to a particular useful application; or

• Other meaningful limitations beyond generally linking the use of the judicial exception to a particular technological environment.

Limitations that were found not to be enough to qualify as "significantly more" when recited in a claim with a judicial exception include:

• Adding the words "apply it" (or an equivalent) with the judicial exception, or mere instructions to implement an abstract idea on a computer;

• Simply appending well-understood, routine and conventional activities previously known to the industry, specified at a high level of generality, to the judicial exception;

• Adding insignificant extrasolution activity to the judicial exception; or

• Generally linking the use of the judicial exception to a particular technological environment or field of use.

A two-step procedure is used to determine if claimed subject matter is a judicial exception. First it is determined whether the claimed subject matter is directed to a law of nature, a natural phenomenon, or an abstract idea. The processes of claims 68-74 and 77-87 are directed to a process of obtaining genomic copy number information from genomic nucleic acids wherein steps e) and f) in independent claim 68 and dependent claims 69-72, 74, and 82 are considered as mental steps. The process of claim 75 is directed to a process of obtaining mRNA copy number information from mRNA transcripts wherein steps d) and e) are considered as mental steps. **Note that steps in a claim that recite the manipulation of information through a series of mental steps would be considered a single abstract idea for purposes of analysis** (see MPEP 2106.04).

In the second step it is determined whether the claimed subject matter includes elements that amount to significantly more than the judicial exception. The claims have additional limitations that constitute conventional steps appended to the abstract ideas. In independent claim 68, the steps a) to d) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see claims 1-21 of US Patent No. 9,670,536 B2 (a priority date of September 21, 2010) and pages 119 and 120, online methods and Figure 1 from Hiatt *et al.,* Nature Methods, 7, 119-122, published online on January 17, 2010) while, in independent claim 75, the steps a) to c) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see Figures 1a, 1c and 1d in page 529 and online methods from Plessy *et al.,* Nature Methods, 7,

528-534, published online on June 13, 2010). The conventional steps do not comprise an inventive concept that transforms the claimed abstract ideas into a patent-eligible application of the abstract ideas. The claimed subject matters of claims 68-75 and 77-87 are processes directed to abstract ideas. When recited at this high level of generality, there is no meaningful limitation, such as a particular or unconventional machine or a transformation of a particular article, in this step that distinguishes it from well-understood, routine, and conventional data gathering activity engaged in by scientists prior to applicant's invention, and at the time the application was filed, *e.g.*, the routine and conventional techniques for performing steps a) to d) of claim 67 or steps a) to c) of claim 75. Further, it is well established that the mere physical or tangible nature of additional elements such as steps a) to d) of claim 67 or steps a) to c) of claim 75 do not automatically confer eligibility on a claim directed to an abstract idea (*see*, *e.g.*, *Alice Corp. v. CLS Bank Int'l*, 134 S. Ct. 2347, 2358-59 (2014)). Therefore, the claimed subject matters of claims 68-75 and 77-87 are without limitations that indicate that the claims are no different from the process claims directed to abstract ideas and are patent-ineligible.

Double Patenting

16. The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. A nonstatutory double patenting rejection is appropriate where the conflicting claims are not identical, but at least one examined application claim is not patentably distinct from the reference claim(s) because the examined application claim is either

anticipated by, or would have been obvious over, the reference claim(s). See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) or 1.321(d) may be used to overcome an actual or provisional rejection based on nonstatutory double patenting provided the reference application or patent either is shown to be commonly owned with the examined application, or claims an invention made as a result of activities undertaken within the scope of a joint research agreement. See MPEP § 717.02 for applications subject to examination under the first inventor to file provisions of the AIA as explained in MPEP § 2159. See MPEP §§ 706.02(l)(1) - 706.02(l)(3) for applications not subject to examination under the first inventor to file provisions of the AIA. A terminal disclaimer must be signed in compliance with 37 CFR 1.321(b).

The USPTO Internet website contains terminal disclaimer forms which may be used. Please visit www.uspto.gov/patent/patents-forms. The filing date of the application in which the form is filed determines what form (e.g., PTO/SB/25, PTO/SB/26, PTO/AIA/25, or PTO/AIA/26) should be used. A web-based eTerminal Disclaimer may be filled out completely online using web-screens. An eTerminal Disclaimer that meets all requirements is auto-processed and approved immediately upon submission. For more information about eTerminal Disclaimers, refer to www.uspto.gov/patents/process/file/efs/guidance/eTD-info-I.jsp.

17. Claims 68-75 and 77-87 are rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 1-20 of U.S. Patent No. 9,404,156 B2. Although the conflicting claims are not identical, they are not patentably distinct from each other because the examined claims in this instant application are either anticipated by, or would have been obvious over, the reference claims. See *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); and, *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969). Although claims 68-75 and 77-87 in this instant application are not identical to claims 1-20 of U.S. Patent No. 9,404,156 B2, claims 1-20 of U.S. Patent No. 9,404,156 B2 are directed to the same subject matter and fall entirely within the scope of claims 68-75 and 77-87 in this instant application. In other words, claims 68-75 and 77-87 in this instant application are anticipated by claims 1-20 of U.S. Patent No. 9,404,156 B2.

Conclusion

18. No claim is allowed.

19. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993)(See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Frank W Lu /
Primary Examiner, Art Unit 1634
April 13, 2018

Notice of References Cited	Application/Control No. 15/063,278	Applicant(s)/Patent Under Reexamination HICKS ET AL.	
	Examiner FRANK LU	Art Unit 1634	Page 1 of 1

U.S. PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Name	CPC Classification	US Classification
*	A	US-9,670,536 B2	06-2017	Casbon; James	C12Q1/6855	1/1
	B	US-				
	C	US-				
	D	US-				
	E	US-				
	F	US-				
	G	US-				
	H	US-				
	I	US-				
	J	US-				
	K	US-				
	L	US-				
	M	US-				

FOREIGN PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Country	Name	CPC Classification
	N					
	O					
	P					
	Q					
	R					
	S					
	T					

NON-PATENT DOCUMENTS

*		Include as applicable: Author, Title Date, Publisher, Edition or Volume, Pertinent Pages)
	U	Hiatt et al., Parallel, tag-directed assembly of locally derived short sequence reads. Nature Methods, 7, 119-122, published online on January 17, 2010.
	V	Plessy et al., Linking promoters to functional transcripts in small samples with nanoCAGE and CAGEscan. Nature Methods, 7, 528-534, published online on June 13, 2010.
	W	
	X	

*A copy of this reference is not being furnished with this Office action. (See MPEP § 707.05(a).)
Dates in MM-YYYY format are publication dates. Classifications may be US or foreign.

GUARDANT HEALTH EX.1009.253

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
INFORMATION DISCLOSURE STATEMENT (Use several sheets if necessary)	**Application Number**	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	9,404,156 B2	08-02-2016	Hicks et al.
	2	9,476,095 B2	10-25-2016	Vogelstein et al.
	3	9,487,829 B2	11-08-2016	Vogelstein et al.
	4	2016/0215333 A1	07-28-2016	Vogelstein et al.
	5	2017/0051347 A1	02-23-2017	Vogelstein et al.

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
Second IDS Exhibit A

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	5,639,603	06-17-1997	Dower et al.
	2	7,537,897	05-26-2009	Brenner et al.
	3	7,622,281	11-24-2009	Ronaghi et al.
	4	8,663,917	03-04-2014	Wigler et al.
	5	8,694,263	04-08-2014	Wigler et al.
	6	6,569,617	05-27-2013	Wigler et al.
	7	7,531,307	05-12-2009	Wigler et al.
	8	8,173,871	09-25-2012	Hannon et al.
	9	8,554,488	10-08-2013	Wigler et al.
	10	7,700,324	04-20-2010	Issa et al.
	11	6,172,214	01-09-2001	Brenner et al.
	12	2004/110153	06-10-2004	Dong et al.
	13	2007/172873	07-26-2007	Brenner et al.
	14	2010/178655	07-15-2010	Hamilton et al.
	15	2006/0073506	04-06-2006	Christians et al.
	16	2005/0032095	02-10-2005	Wigler et al.
	17	2005/0266444	12-01-2005	Wigler et al.
	18	2007/0207481	09-06-2007	Wigler et al.
	19	2010/0227768	09-10-2010	Wigler et al.
	20	2012/0149593	06-14-2012	Hicks et al.
	21	2012/0328607	12-27-2012	Hicks et al.
	22	2013/0179999	07-11-2013	Hannon et al.

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
First IDS Exhibit A

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
INFORMATION DISCLOSURE STATEMENT (Use several sheets if necessary)	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	23	2014/0025307	01-23-2014	Wigler et al.
	24	2006/0035258	02-16-2006	Tadakamalla et al.
	25	2009/0137402	05-28-2009	Wang et al.
	26	2010/0203538	08-12-2010	Dube et al.

FOREIGN PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Foreign Patent Document Country Code[3] Number[4] Kind Code[5] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document	T[6]
	27	WO 2007/062445	06-07-2007	Alexander Seitz	
	28	WO 2012/054873	04-26-2012	Hicks et al.	

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
(Use several sheets if necessary)	Attorney Docket No.	81503-AA/GJG/AWG/AC

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	29	February 13, 2014 Extended European Search Report, issued in connection with European Patent Application No. 11835243.4	
	30	April 23, 2013 International Preliminary Report on Patentability issued in connection with PCT International Application No. PCT/US2011/057350 (WO 2012/054873)	
	31	June 26, 2012 International Search Report issued in connection wth PCT International Application No. PCT/US2011/057350 (WO 2012/054873)	
	32	Miner et al., "Molecular barcodes detect redundancy and contamination in hairpin-bisulfite PCR," Nucleic Acids Res. vol. 32 no. 17 (2004), p. 1-4	
	33	Casbon et al., "A method for counting PCT template molecules with application to next-generation sequencing", Nucleic Acids Res. vol. 39 no. 12 (July 2011)	
	34	Cheung V.G. & Nelson S.F., "Whole genome amplification using a degenerate oligonucleotide primer allows hundreds of genotypes to be performed on less than one nanogram of genomic DNA," PNAS vol. 93 no. 25 (Dec. 1996), p. 14676–79	
	35	Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," Science vol. 323 no. 5910 (Jan. 2009), p. 133-38	
	36	Supporting Online Material for Eid et al., "Real-Time DNA Sequencing from Single Polymerase Molecules," accessible at www.sciencemag.org/cgi/content/full/1162986/DC1, (published Nov. 20, 2008)	
	37	Fu G.K. et al., "Counting individual DNA molecules by the stochastic attachment of diverse labels," PNAS vol. 108 no. 22 (May 2011), p. 9026-31	
	38	McCloskey M.L. et al, "Encoding PCR Products with Batch-stamps and Barcodes," Biochem Genet. vol. 45 (Dec. 2007), p. 761-67	
	39	Parameswaran et al., "A pyrosequencing-tailored nucleotide barcode design unveils opportunities for large-scale sample multiplexing," Nucleic Acids Res. vol. 35 no. 19 (2007)	
	40	Paunio T. et al., "Preimplantation diagnosis by whole genome amplification, PCT amplification, and solid-phase minisequencing of blastomere DNA," Clinical Chem. vol. 42 no. 9 (1996), p. 1382-90	
	41	Technology Backgrounder: Single Molecule Real Time (SMRT™) DNA Sequencing, Pacific Bioscience (2009)	
	42	Islam et al., "Characterization of the single-cell transcriptional landscape by highly multiplex RNA-seq," Genome Res. vol. 21 no. 7 (July 2011) p. 1160-67	

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
	Application Number	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
INFORMATION DISCLOSURE STATEMENT	Art Unit	1634
(Use several sheets if necessary)	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/AWG/AC

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	43	Kivioja et al., "Counting absolute numbers of molecules using unique molecular identifiers, Nature Methods vol. 9 (2012), p. 72-74	
	44	Shiroguchi at al., "Digital RNA sequencing minimizes sequence-dependent bias and amplification noise with optimized single-molecule barcodes," PNAS vol. 109 no. 4 (2012), p. 1347-52 ,	
	45	Castle et al., "DNA copy number, including telomeres and mitochondria, assayed using next-generation sequencing," BMC Genomics vol. 11 no. 244 (2010)	
	46	Craig et al., "Identification of Genetic Variants Using Bar-coded Multiplexed Sequencing," Nature Methods vol. 5 no. 10 (Oct. 2008), p. 887-93.	

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15063278	HICKS ET AL.
	Examiner	Art Unit
	FRANK LU	1634

CPC- SEARCHED		
Symbol	Date	Examiner
See attached file	4/13/2018	/FL/

CPC COMBINATION SETS - SEARCHED		
Symbol	Date	Examiner
See attached file	4/13/2018	/FL/

US CLASSIFICATION SEARCHED			
Class	Subclass	Date	Examiner
435	6.1, 6.11, 6.12, 91.1, 91.2, 183	4/13/2018	/FL/
436	94, 501	4/13/2018	/FL/
536	23.1, 24.3, 24.33, 25.3	4/13/2018	/FL/

* See search history printout included with this form or the SEARCH NOTES box below to determine the scope of the search.

SEARCH NOTES		
Search Notes	Date	Examiner
STN and EAST (see attached file) and Inventor Searches	4/13/2018	/FL/

INTERFERENCE SEARCH			
US Class/ CPC Symbol	US Subclass / CPC Group	Date	Examiner

	/FRANK LU/ Primary Examiner.Art Unit 1634

The similar searches were performed in STN.

EAST Search History

EAST Search History (Prior Art)

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	31	(tag or adaptor or adapter or linker) same ("nucleic acid" or polynucleotide or DNA) same (PCR near (polymerase near chain near reaction)) same sequence near read	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 20:53
L2	581	(tag or adaptor or adapter or linker) same ("nucleic acid" or polynucleotide or DNA) same (amplify or amplifying or amplification) same sequence near read	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 20:59
L3	298	(tag or adaptor or adapter or linker) same ("nucleic acid" or polynucleotide or DNA) same (amplify or amplifying or amplification) same sequence near read and (PCR near (polymerase near chain near reaction))	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 21:00
L4	274	(tag or adaptor or adapter or linker) same ("nucleic acid" or polynucleotide or DNA) same (amplify or amplifying or amplification) same sequence near read and (PCR near (polymerase near chain near reaction))same (amplify or amplifying or amplification)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 21:00
L5	233	(tag or adaptor or adapter or linker) near10("nucleic acid" or polynucleotide or DNA) same (amplify or amplifying or amplification) same sequence near read and (PCR near (polymerase near chain near reaction))same (amplify or amplifying or amplification)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 21:01
L6	41	(tag or adaptor or adapter or linker) near10("nucleic acid" or polynucleotide or DNA) near20 (amplify or amplifying or amplification) near20 sequence near read and (PCR near (polymerase near chain near reaction))same (amplify or amplifying or amplification)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 21:01
L13	39	l12 and sequence near read	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	ON	2018/04/13 21:09
L14	41	(tag or adaptor or adapter or linker)	US-PGPUB;	OR	ON	2018/04/13

		near10("nucleic acid" or polynucleotide or DNA) near20 (amplify or amplifying or amplification) near20 sequence near reads and (PCR near (polymerase near chain near reaction))same (amplify or amplifying or amplification)	USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB			21:39

4/ 13/ 2018 10:09:06 PM
C:\ Users\ flu\ Documents\ EAST\ Workspaces\ ~ auto.05052017.131049.bak.wsp

Form PTO-1449 (Substitute) U.S. Department of Commerce Patent and Trademark Office		
INFORMATION DISCLOSURE STATEMENT (Use several sheets if necessary)	**Application Number**	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Wei Min Lu
	Attorney Docket No.	81503-AA/GJG/ADR/AMM

U.S. PATENT DOCUMENTS

Examiner Initials*	Cite No.[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM-DD-YYYY	Name of Patentee or Applicant of Cited Document
	1	6,013,445	01-11-2000	Albrecht et al.

NON PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No.[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.	T[2]
	2	October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	3	February 16, 2018 Amendment under Rule 71(3) EPC to October 9, 2017 Communication under Rule 71(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	4	December 8, 2017 Office Action issued by the Canadian Intellectual Property Office in connection with Canadian Patent Application 2,815,076	
	5	May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807	
	6	April 19, 2017 Response to May 3, 2016 Patent Examination Report No. 1 issued by IP Australia in connection with Australian Patent Application 2011316807	
	7	April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807	
	8	May 2, 2017 Response to April 20, 2017 Patent Examination Report No. 2 issued by IP Australia in connection with Australian Patent Application 2011316807	
	9	September 7, 2017 Notice of Grant for Patent issued by IP Australia in connection with Australian Patent Application 2011316807	
	10	November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	
	11	June 27, 2016 Amendment under Rule 94(3) EPC to November 20, 2015 Communication under Rule 94(3) EPC issued by the European Patent Office in connection with European Patent Application No. 11835243.4	

EXAMINER SIGNATURE	/FRANK W LU/	DATE CONSIDERED	04/13/2018

*EXAMINER: Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1]Applicant's unique citation designation number (optional). [2]Applicant is to place a checkmark here if English language Translation is attached.

Applicants: James Hicks
Serial No.: 15/063,278
Filing Date: March 7, 2016
Third IDS Exhibit A

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L./

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☒ SMALL ☐ MICRO

APPLICATION AS FILED – PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA	RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A	N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A	N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A	N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*	X $ =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*	X $ =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).			
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))				
* If the difference in column 1 is less than zero, enter "0" in column 2.			TOTAL	

APPLICATION AS AMENDED – PART II

AMENDMENT **10/17/2018**

	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0	X $50 =	0
Independent (37 CFR 1.16(h))	* 2	Minus	***3	= 0	X $230 =	0
☐ Application Size Fee (37 CFR 1.16(s))						
☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
					TOTAL ADD'L FEE	0

AMENDMENT

	(Column 1) CLAIMS REMAINING AFTER AMENDMENT		(Column 2) HIGHEST NUMBER PREVIOUSLY PAID FOR	(Column 3) PRESENT EXTRA	RATE ($)	ADDITIONAL FEE ($)
Total (37 CFR 1.16(i))	*	Minus	**	=	X $ =	
Independent (37 CFR 1.16(h))	*	Minus	***	=	X $ =	
☐ Application Size Fee (37 CFR 1.16(s))						
☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))						
					TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.

** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".

*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".

The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.

LIE
VICTOR BARLOW

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

DocCode – SCORE

SCORE Placeholder Sheet for IFW Content

Application Number: 15063278 Document Date: 10/17/2018

The presence of this form in the IFW record indicates that the following document type was received in electronic format on the date identified above. This content is stored in the SCORE database.

Since this was an electronic submission, there is no physical artifact folder, no artifact folder is recorded in PALM, and no paper documents or physical media exist. The TIFF images in the IFW record were created from the original documents that are stored in SCORE.

- Drawing

At the time of document entry (noted above):

- USPTO employees may access SCORE content via eDAN using the Supplemental Content tab, or via the SCORE web page.
- External customers may access SCORE content via PAIR using the Supplemental Content tab.

Form Revision Date: August 26, 2013

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 10/17/2018

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
10/17/2018	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.265

UNITED STATES DEPARTMENT OF COMMERCE
U.S. Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450

APPLICATION NO./ CONTROL NO.	FILING DATE	FIRST NAMED INVENTOR/ PATENT IN REEXAMINATION	ATTORNEY DOCKET NO.
15/063,278	03/07/2016	Hicks et al.	81503-AA/GJG/AWG/

	EXAMINER	
COOPER & DUNHAM, LLP 30 Rockefeller Plaza 20th Floor NEW YORK, NY 10112	FRANK LU	
	ART UNIT	PAPER
	1634	20181015

DATE MAILED: _____

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner for Patents

This is a decision on the Petition to Accept Color Drawings under 37 C.F.R 1.84, received in the United States Patent and Trademark Office (USPTO) on March 7, 2016.

A review of U.S. Patent and Trademark records indicates that all of the requirements for acceptance of color drawings or photographs have been met.

Accordingly, the petition under 37 C.F.R. 1.84 is GRANTED.

Should there be any questions with respect to this decision, please contact:
Scarlett Goon (571-270-5241)
TC Quality Assurance Specialist
TC1600

/SCARLETT Y GOON/ Quality Assurance Specialist, TC 1600	/Zachariah Lucas/ Supervisory Patent Examiner, Art Unit 1600

PTO-90C (Rev.04-03)

Technology Center Petition Count Sheet

Application No.	Applicant(s)	
15/063,278	Hicks et al.	

Petition Code	Petition Type
☐ 601	Petition to Make Special, Age or Health (37 CFR 1.102, MPEP § 708.02)
☐ 604	Petition to Make Special, Accelerated Examination (37 CFR 1.102, MPEP § 708.02)
☐ 607	Petition to Make Special, For Reasons not Provided in 601-606 and 639-640 (37 CFR 1.102, MPEP § 708.02)
☐ 608	Petition to Reopen After a Board Decision (37 CFR 1.198)
☐ 609	Petition for Review of Final Restriction Requirement (37 CFR 1.144)
☐ 610	Petition Invoking Authority of Commissioner under 37 CFR 1.181 Not Specified in Codes 611-617
☐ 611	Petition Relating to Prematureness of Final Rejection (37 CFR 1.181, MPEP § 706.07)
☐ 612	Petition Relating to Refusal to Enter an Amendment (37 CFR 1.181 and 1.127, MPEP § 714.09)
☐ 613	Petition Relating to Withdraw of holding of Abandonment (37 CFR 1.137, MPEP § 711.03)
☐ 614	Petition Relating to Withdraw of holding of Abandonment (37 CFR 1.137, MPEP § 7.14.19)
☐ 615	Petition Relating to Formal Sufficiency / Propriety of Affidavits (37 CFR 1.131, 1.132, 1.608, MPEP § 608.04(C)
☐ 617	Petition Relating to Refusal to Enter an Amendment (37 CFR 1.312)
☐ 620	Petition for Extension of Time (37 CFR 1.136 (b))
☐ 622	Petition Concerning Appeal Application before Transfer of Jurisdiction to Board of Appeals (MPEP § 1206)
☐ 623	Petition for Second or Subsequent Suspension of Action (37 CFR 1.103, MPEP § 709)
☐ 634	Petition to Correct Inventorship in a Patent not in Interference (37 CFR 1.324, MPEP § 1481)
☐ 635	Petition to Correct Inventorship in a Patent Application (37 CFR 1.48)
☐ 636	Petition to Change Inventorship in a Patent (37 CFR 1.324)
☐ 637	Petition to Withdraw from Issue, before Payment of an Issue Fee (37 CFR 1.313(a))
☐ 639	Petition for Make Special Prospective Manufacture (37 CFR 1.102, MPEP § 708.02)
☐ 646	Petition for Make Special Accelerated Examination
☑ 690	Petition Related Color Drawings of Photographs
☐ 699	Petition for Matters Before Technology Center

Information Required for OPESS Processing

☑ GRANTED ☐ GRANTED-IN-PART ☐ DENIED ☐ DISMISSED

☑ Mail Decision and Enter Decision in Petition Database/PALM
☐ Copy Decision, Send to Scanning
☐ Fees Need to be Charged
☐ _____

GUARDANT HEALTH EX.1009.267

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
October 17, 2018

BY EFS
Commissioner for Patents
Alexandria, VA

AMENDMENT IN RESPONSE TO APRIL 17, 2018 OFFICE ACTION AND PETITION FOR A THREE-MONTH EXTENSION OF TIME

This Amendment is submitted in response to an April 17, 2018 Office Action issued by the United States Patent and Trademark Office in connection with the above-identified application. A response to the April 17, 2018 Office Action was originally due July 17, 2018. Applicants hereby petition for a three-month extension of time. The required fee for a three-month extension of time is SEVEN HUNDRED DOLLARS ($700.00) for a small entity and authorization is hereby given to charge this fee to Deposit Account No. 03-3125. With a three-month extension of time, a response to the April 17, 2018 Office Action is now due October 17, 2018. Accordingly, this Amendment is being timely filed.

Amendments to the Drawings are on page **2** of this paper.

Amendments to the Specification begin on page **3** of this paper.

Amendments to the Claims begin on page **4** of this paper.

Remarks begin on page **12** of this paper.

AMENDMENTS TO THE DRAWINGS

Applicants attach hereto as **Exhibit A** EIGHT (8) replacement sheets of drawings containing Figures 7-10. Additionally, applicants attach as **Exhibit B**, FOUR (4) sheets of drawings containing previous Figures 7-10, marked to show the amendments.

AMENDMENTS TO THE SPECIFICATION

Please amend the paragraph that runs from lines 4-10 on page 1 of the subject application as previously amended in applicants March 8, 2016 Supplemental Preliminary Amendment, as follows:

--This application is a continuation of U.S. Serial No. 13/278,333, filed October 21, 2011, now ~~allowed,~~ U.S. Patent No. 9,404,156, issued August 2, 2016, which claims the benefit of U.S. Provisional Application Nos. 61/510,579, filed July 22, 2011, and 61/406,067, filed October 22, 2010 the contents of each of which are hereby incorporated by reference in their entirety into this application.

Please amend the paragraph that runs from lines 27-29 on page 13 of the subject application as filed, as follows:

--Figure 5. ROMA CGH of sKBR3 compared with profiles of 3 independent single cell isolates derived from sparse DNA sequencing. (A) Batch DNA. (B, C, D) Single Cell.--

Please amend the paragraph that runs from lines 3-4 on page 14 of the subject application as filed, as follows:

--Figure 7(A-C). Flowchart for genomic profiling by representational sequence counting--

Please amend the paragraph on line 8 of page 14 of the subject application as filed, as follows:

-- Figure 9(A and B). Nick ligation protocol--

Please amend the paragraph on line 15 of page 14 of the subject application as filed, as follows:

--Figure 10(A and B). Schema for targeted sequencing of regional markers--

AMENDMENTS TO THE CLAIMS

Pursuant to 37 C.F.R. § 1.121(c), this listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

1-67. (Canceled)

68. (Currently amended) A method for obtaining genomic copy number information from genomic nucleic acids, comprising:

a) obtaining segments of the genomic nucleic acids;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each of the tag associated sequence read of step (d); and

f) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome thereby obtaining a count for each location on the genome,

thereby obtaining genomic copy number information from the genomic nucleic acids.

69. (Currently amended) The method of claim [[72]]68, further comprising estimating a genomic copy number of ~~a region of the genome comprising~~ multiple locations on the genome within a region of the genome.

70. (Currently amended) The method of claim [[72]]68, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

71. (Currently amended) The method of claim [[72]]68, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

72. (Currently amended) The method of claim [[75]]71, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (h) and the counts obtained in step (j).

73. (Currently amended) The method of claim [[75]]71, wherein the second region of the genome comprises a centromere.

74. (Currently amended) The method of claim [[72]]68, further comprising l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative genomic copy number of the region of the genome.

75. (Previously presented) A method for obtaining mRNA copy number information from mRNA transcripts, comprising:

a) Generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts

under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence

reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library,

thereby obtaining mRNA copy number information from the mRNA transcripts.

76. (Withdrawn - Currently amended) The method of claim [[72]]68, wherein said tagging the segments comprises:

n) adding a polynucleotide tail to the ends of the segments of the genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail;

o) subjecting the zero-th order derivative strands of step (n) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement of the zero-th order derivative strands, thereby generating first order derivative strands;

p) adding a polynucleotide tail to the first order derivative strands, thereby generating a first order derivative strands comprising the polynucleotide tail;

q) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order

derivative strands,

wherein the primers of at least one of steps o) and q) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules.

77. (Currently amended) The method of claim [[72]]68, wherein said tagging the segments comprises:

r) adding a polynucleotide tail to the ends of the segments of genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail,

s) subjecting the zero-th order derivative strands of step (r) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (r) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

t) subjecting the product of step (s) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step r) under conditions which promote the formation of only one complement of the product of step (s), wherein the primers of step (s) and the primers of step (t) have different nucleotide sequences, and wherein the polymerase has 3′-5′ proofreading activity,

wherein the primers of at least one of steps (s) and (t) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, thereby generating unique tagged nucleic acid molecules.

78. (Currently amended) The method of claim [[81]]77, wherein said adding the polynucleotide tail comprises the use of a terminal transferase.

79. (Currently amended) The method of claim [[72]]68, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to step (d).

80. (Currently amended) The method of claim [[72]]68, wherein the tags further comprise a sample tag.

81. (Currently amended) The method of claim [[84]]80, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules product of step (c).

82. (Currently amended) The method of claim [[85]]81, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

83. (Currently amended) The method of claim [[72]]68, wherein the unique tagged nucleic acid molecules are generated from a single cell.

84. (Currently amended) The method of claim [[83]]79, wherein the unique tagged nucleic acid molecules are subjected to hybrid capture prior to the PCR in step (c).

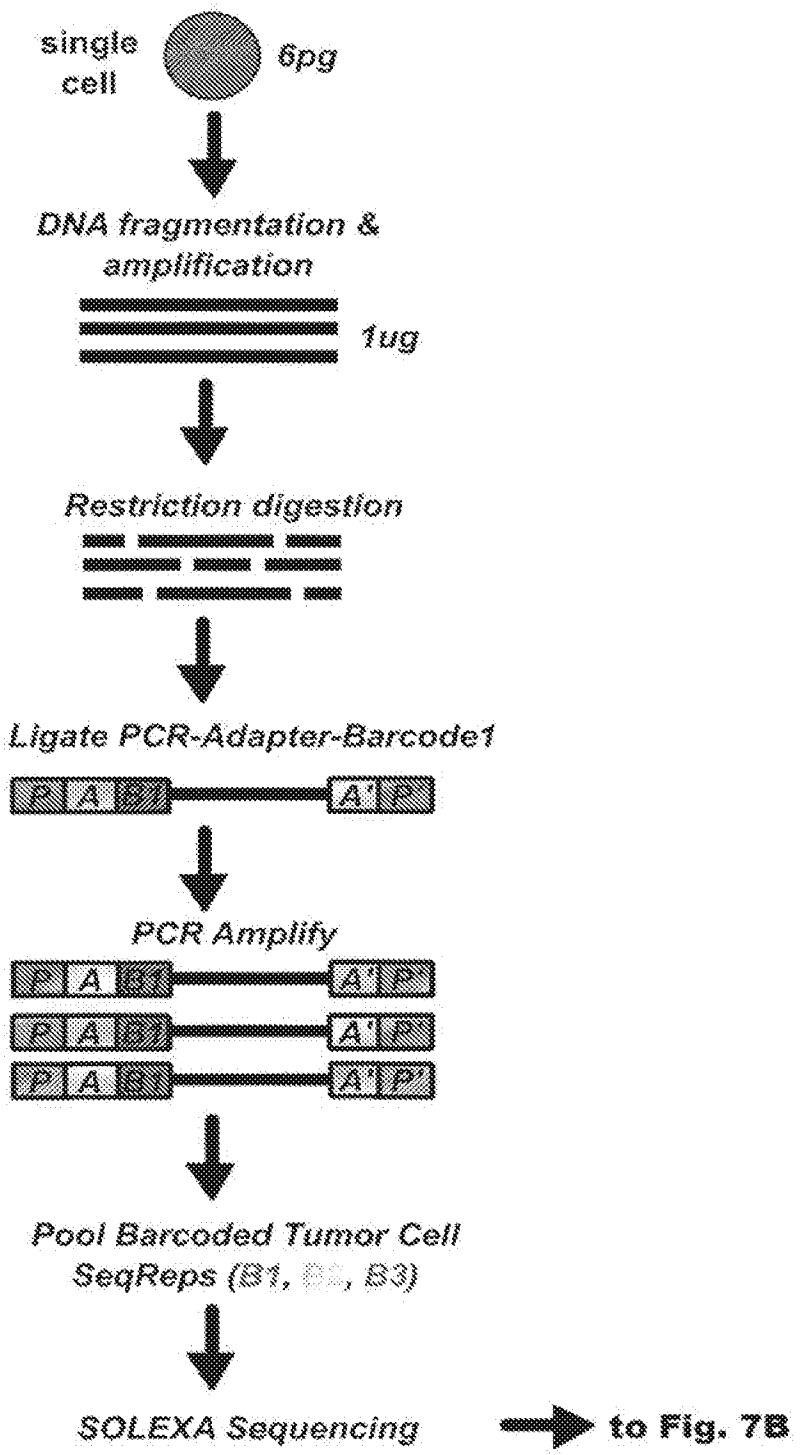
85. (Currently amended) The method of claim [[88]]84, wherein each of the unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array.

86. (Currently amended) The method of claim [[83]]79, wherein the amplified unique tagged nucleic acid molecules produced in step (c) are subjected to hybrid capture prior to said sequencing the

amplified unique tagged nucleic acid molecules in step (d).

87. (Currently amended) The method of claim [[90]]86, wherein each of the amplified unique tagged nucleic acid molecules subject to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on a microarray.

Fig. 7A



U.S. Serial No.: 15/063,278
Applicant(s): James Hicks et al.
Filed: March 7, 2016
Exhibit A

Fig. 7B (continued from Fig. 7A)

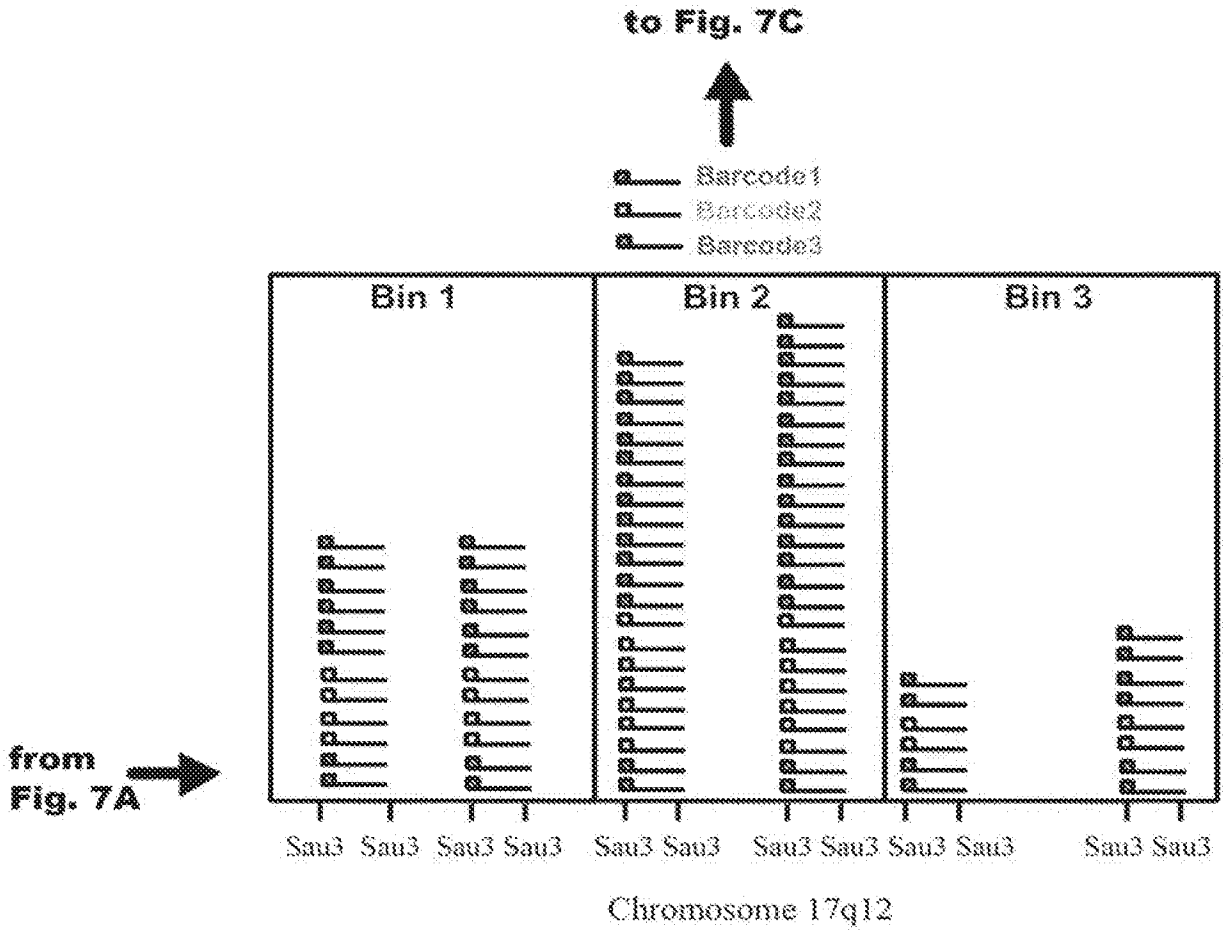
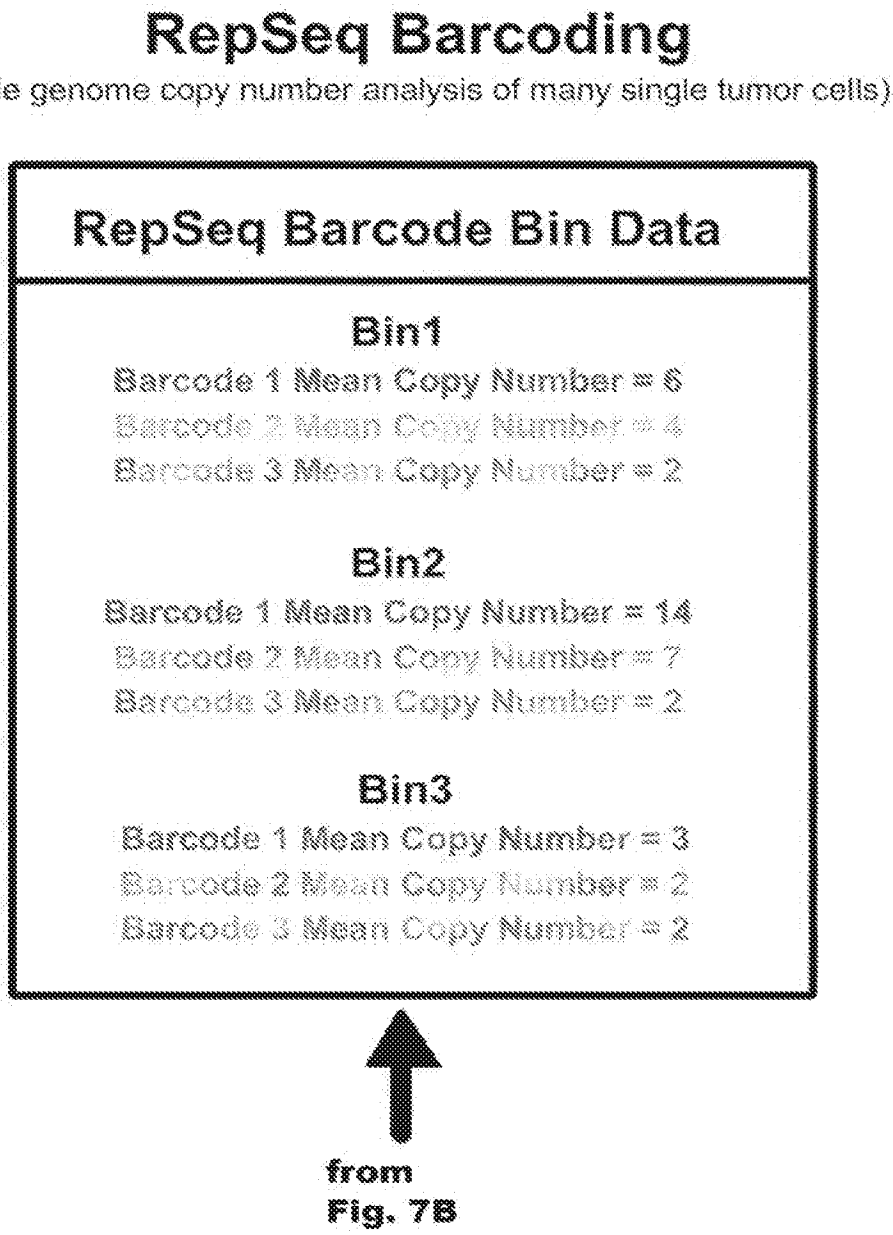


Fig. 7C (continued from Fig. 7B)



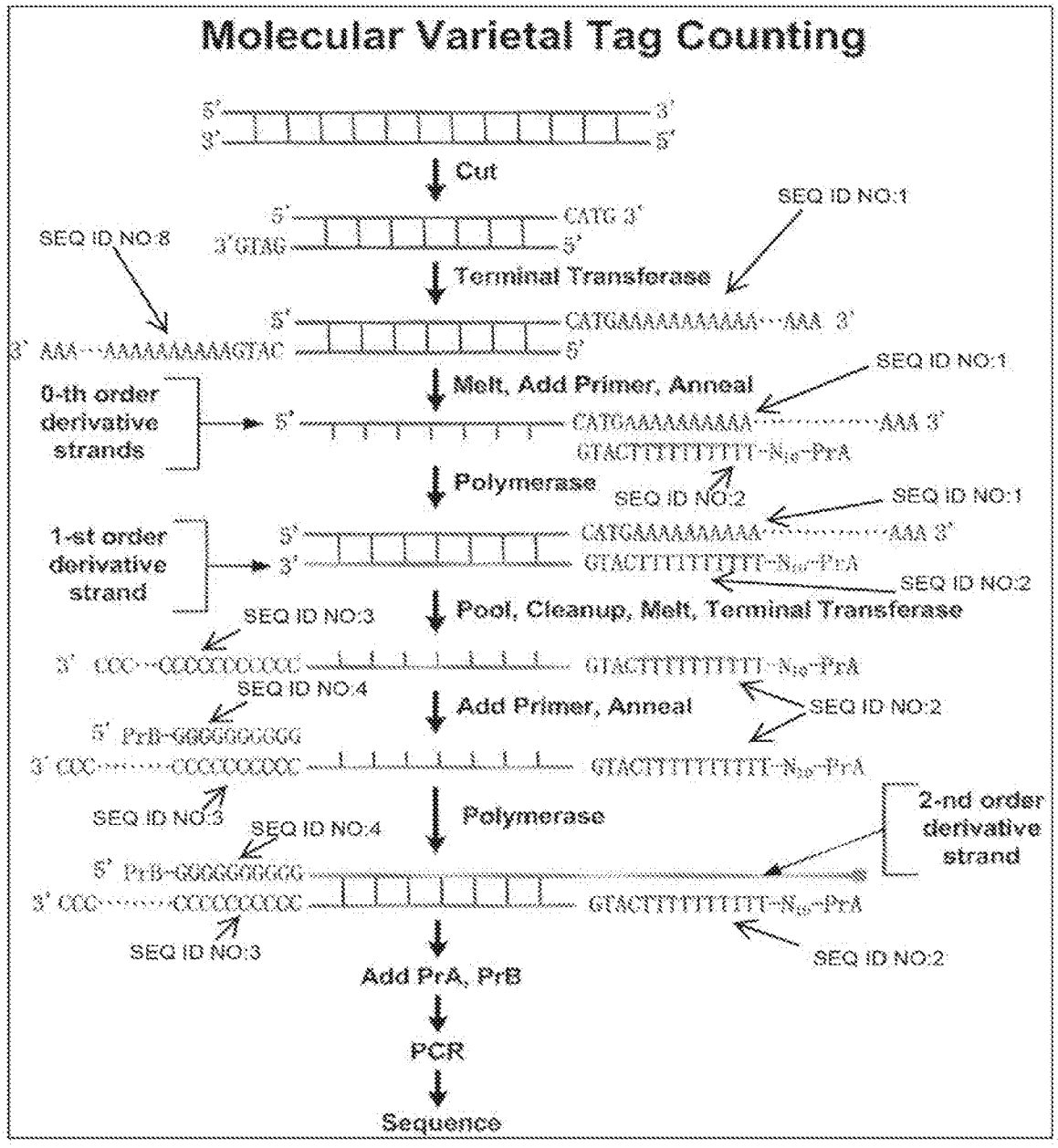
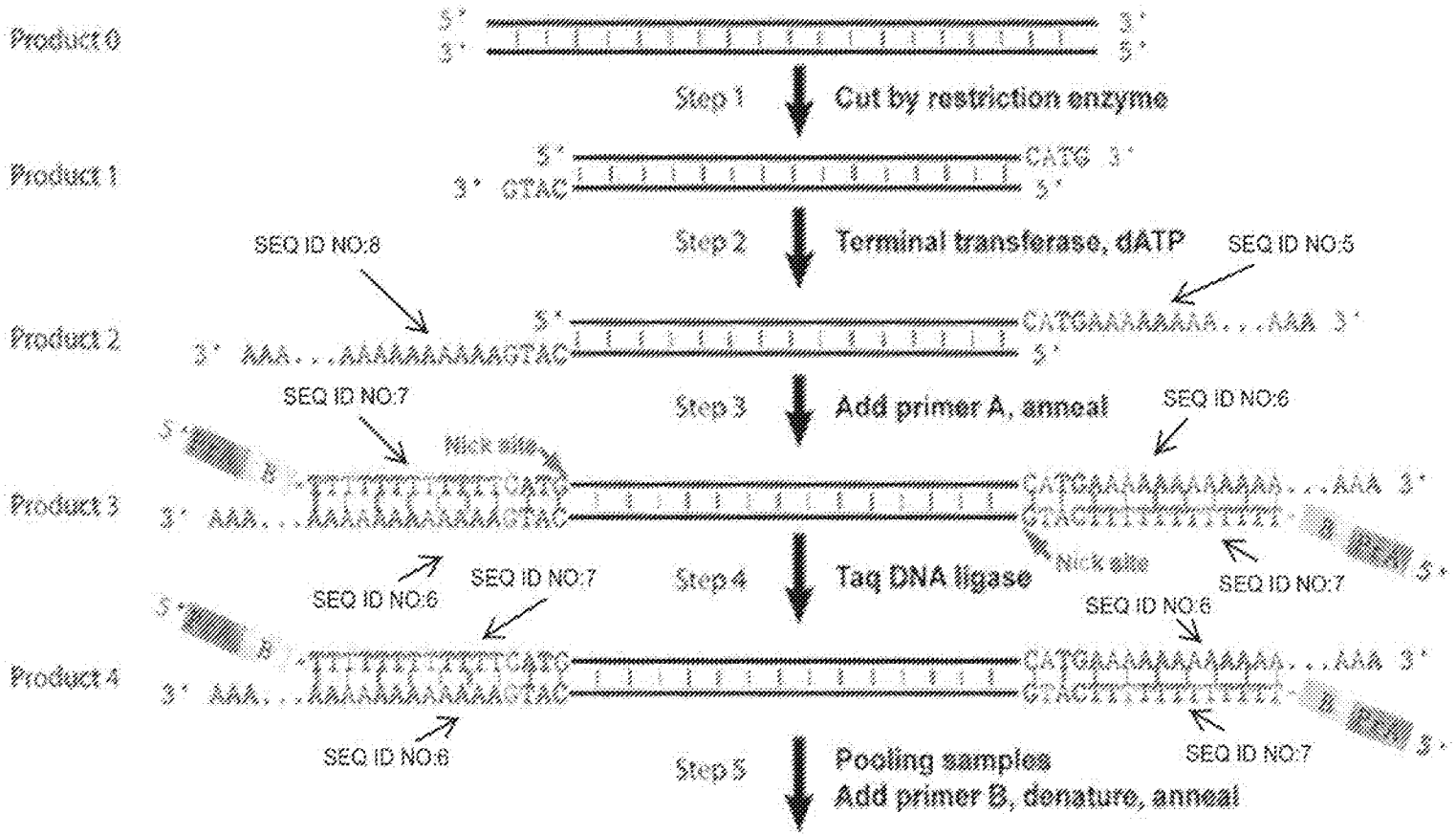

Molecular Varietal Tag Counting
Cut
SEQ ID NO:8
SEQ ID NO:1
Terminal Transferase
Melt, Add Primer, Anneal
0-th order derivative strands
Polymerase
SEQ ID NO:2
1-st order derivative strand
Pool, Cleanup, Melt, Terminal Transferase
SEQ ID NO:3
SEQ ID NO:4
Add Primer, Anneal
2-nd order derivative strand
Add PrA, PrB
PCR
Sequence

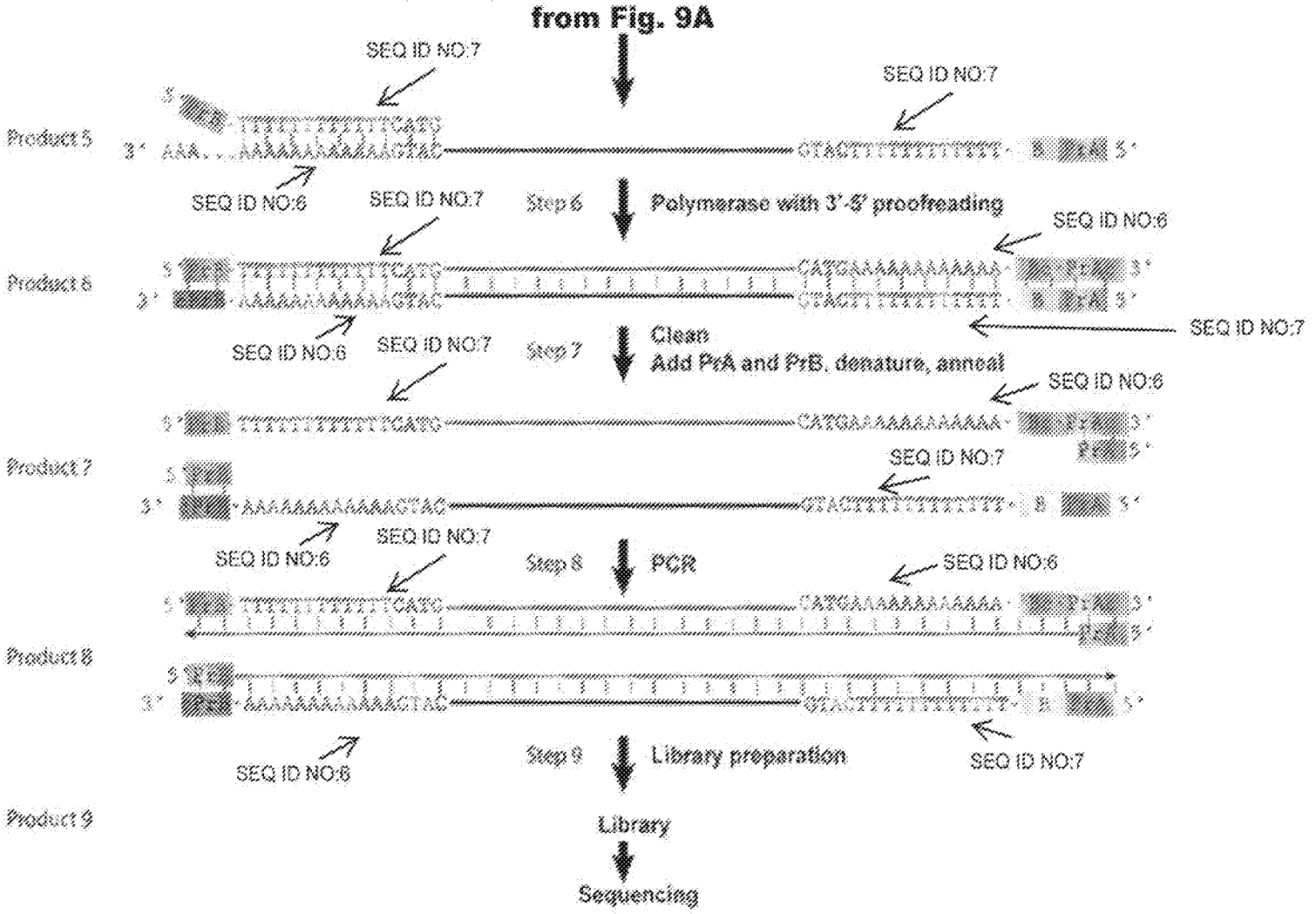
Fig. 8

Fig. 9A



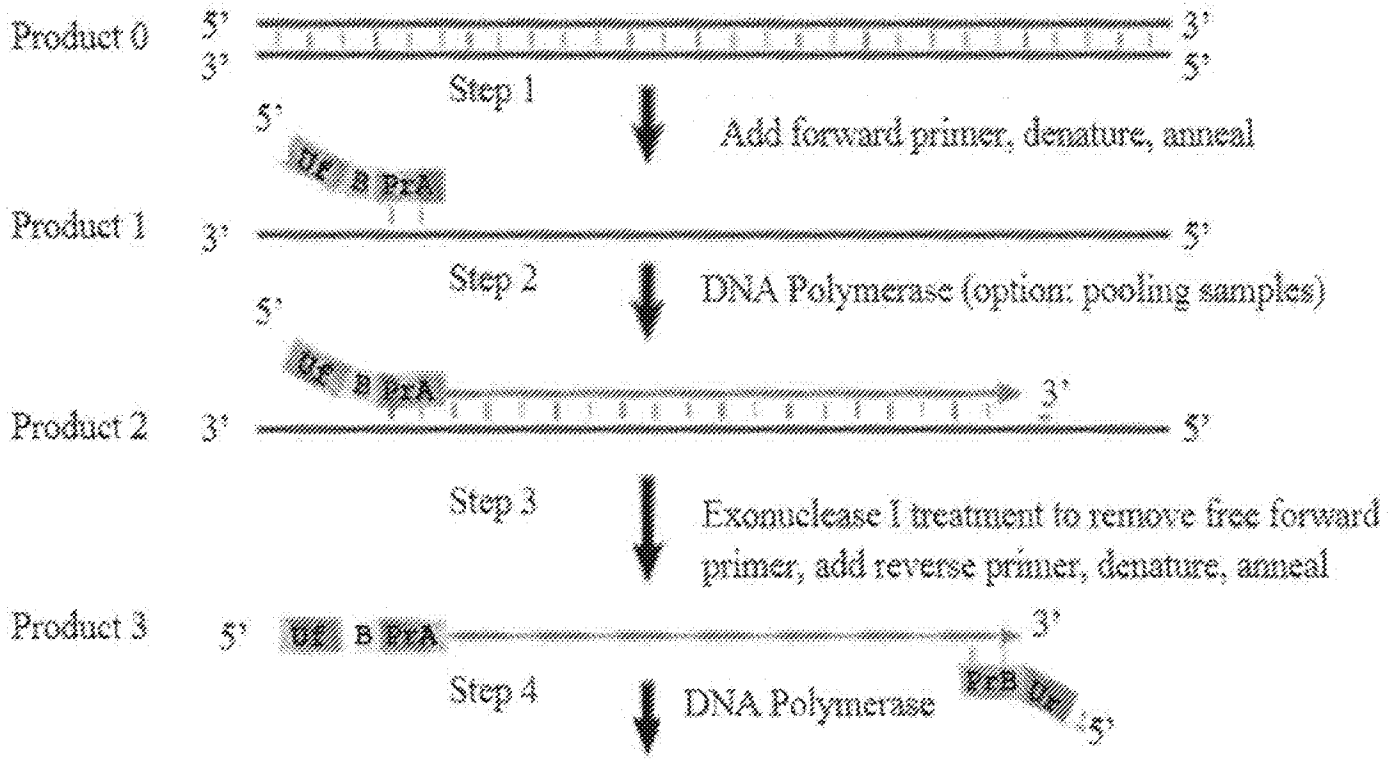
to Fig. 9B

Fig. 9B (continued from Fig. 9A)



GUARDANT HEALTH EX.1009.284

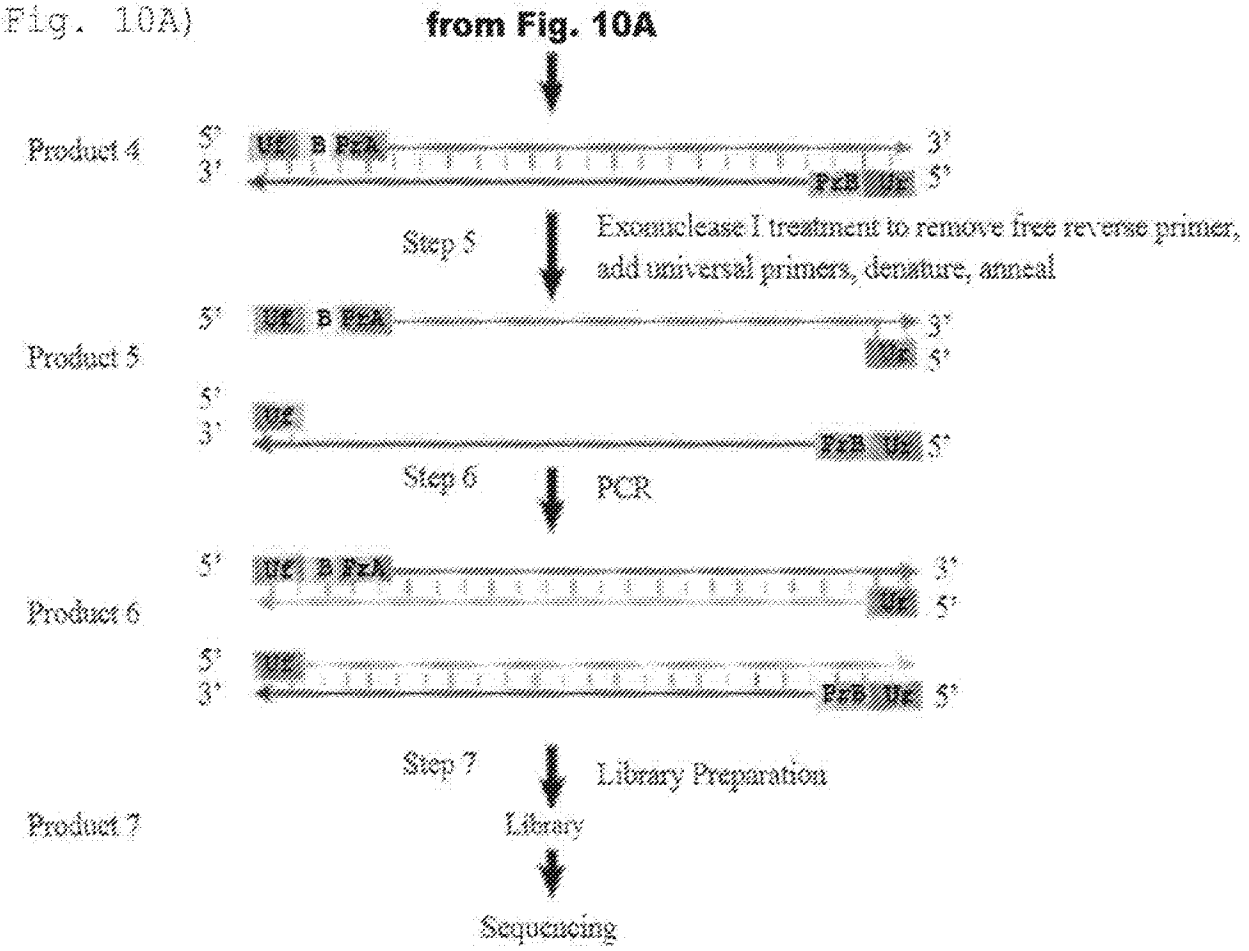
Fig. 10A



to Fig. 10B

Fig. 10B

(continued from Fig. 10A)



GUARDANT HEALTH EX.1009.286

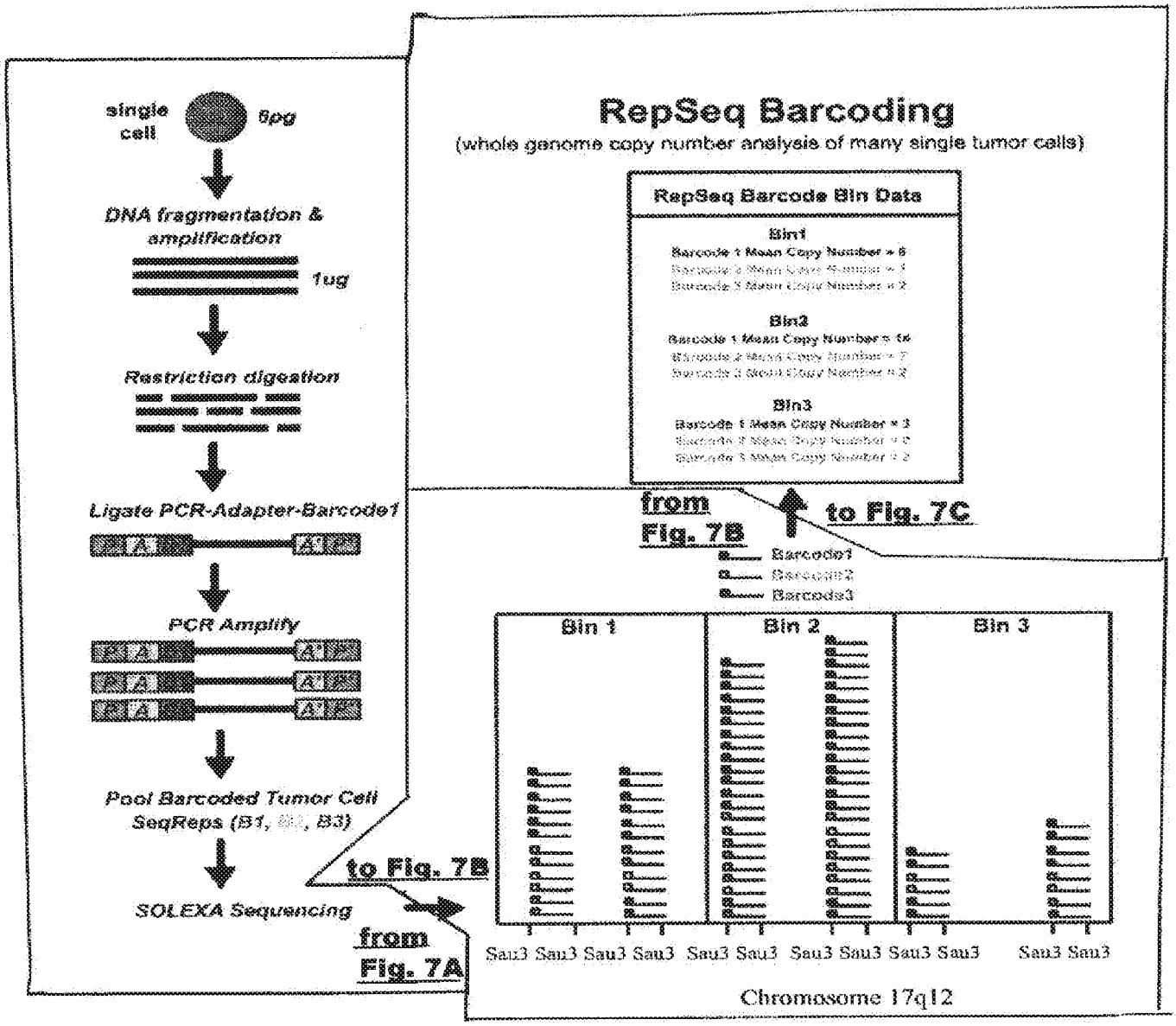


Fig. 7

split into 3 sheets

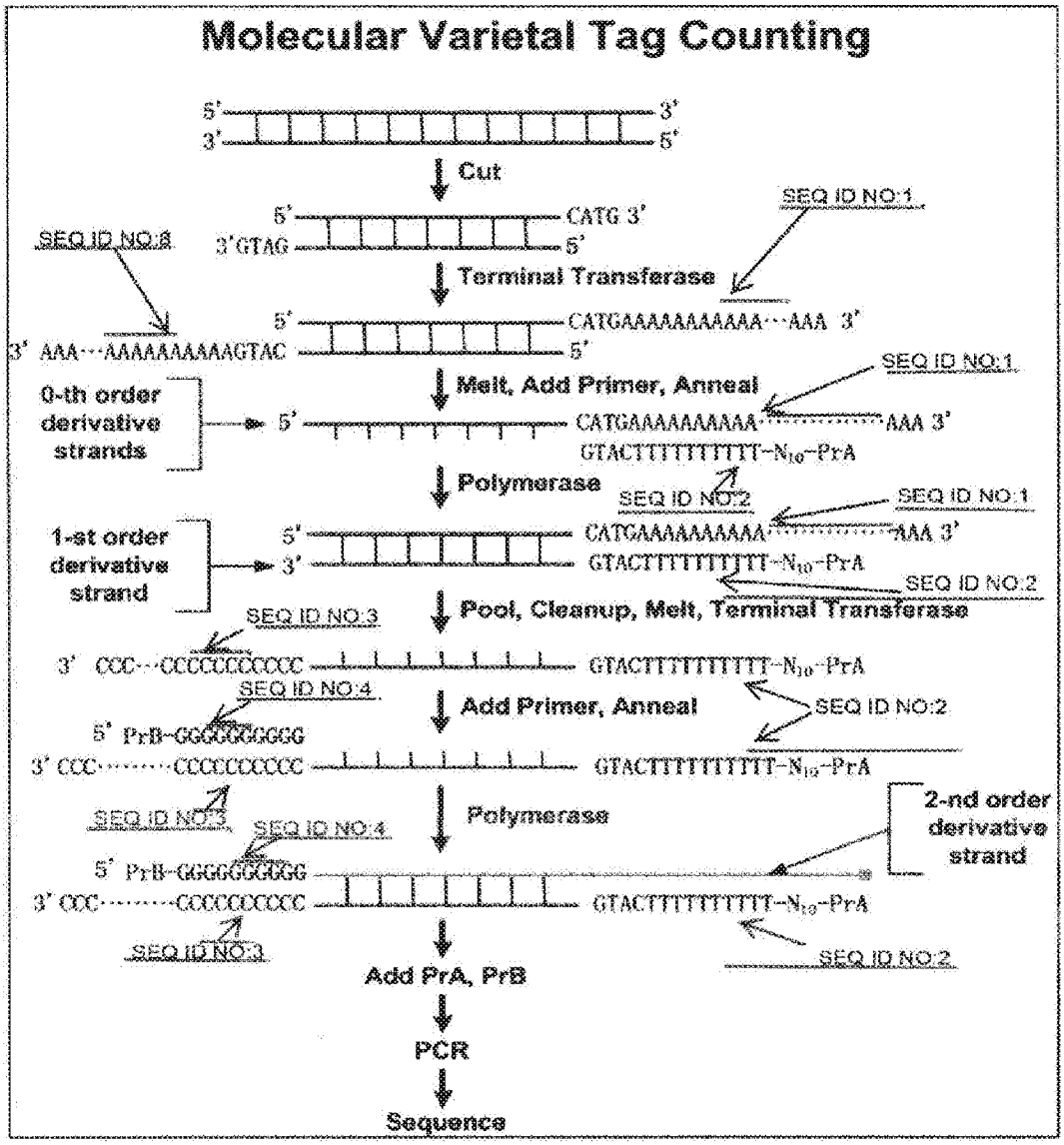

Molecular Varietal Tag Counting
Cut
Terminal Transferase
SEQ ID NO:1
SEQ ID NO:8
Melt, Add Primer, Anneal
0-th order derivative strands
Polymerase
1-st order derivative strand
SEQ ID NO:2
SEQ ID NO:3
Pool, Cleanup, Melt, Terminal Transferase
SEQ ID NO:4
Add Primer, Anneal
2-nd order derivative strand
Add PrA, PrB
PCR
Sequence

Fig. 8

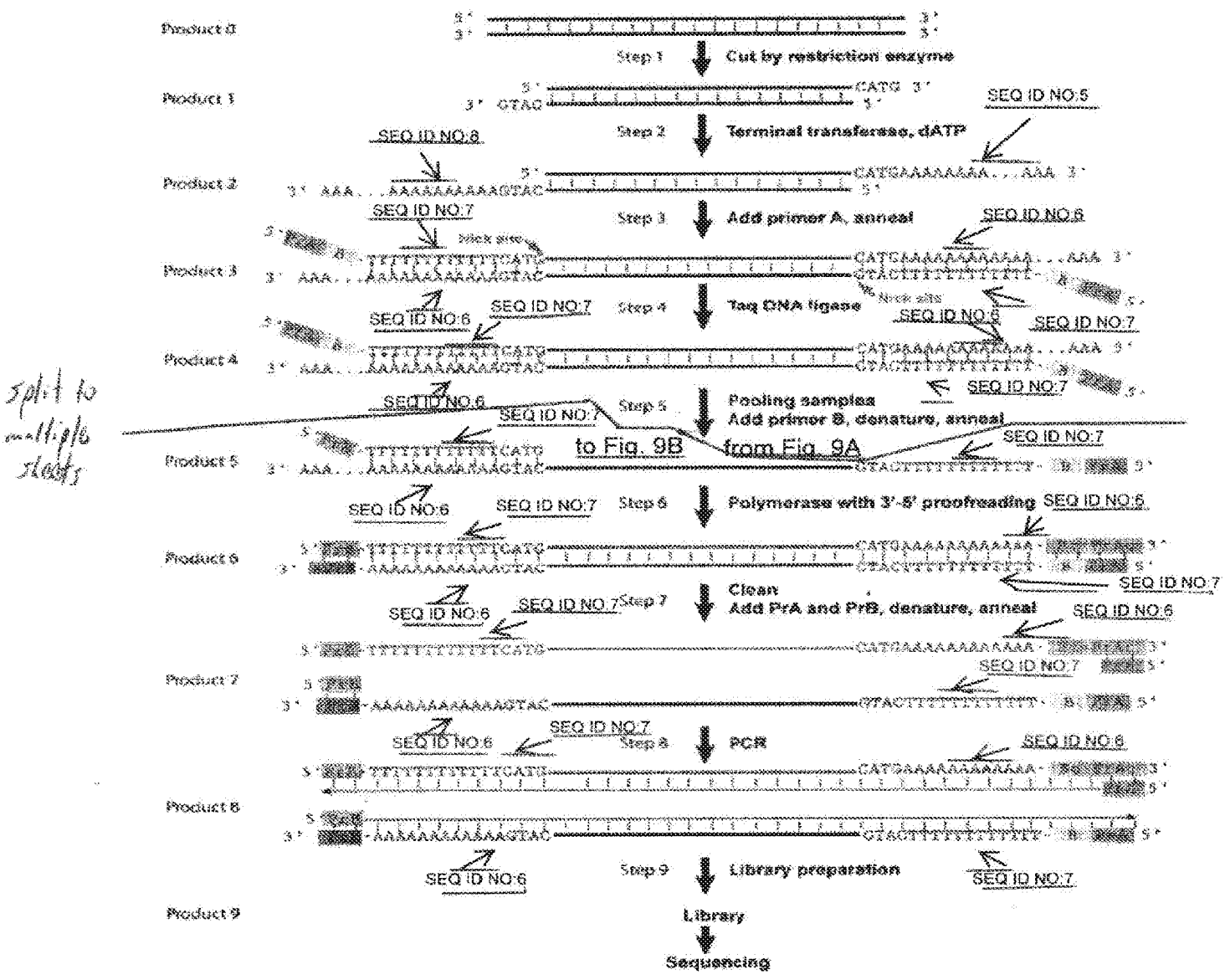

Product 0
Step 1
Cut by restriction enzyme
Product 1
SEQ ID NO:5
SEQ ID NO:8
Step 2
Terminal transferase, dATP
Product 2
SEQ ID NO:7
Step 3
Add primer A, anneal
SEQ ID NO:8
Nick site
Product 3
SEQ ID NO:6
SEQ ID NO:7
Step 4
Taq DNA ligase
SEQ ID NO:8
SEQ ID NO:7
Product 4
Split to multiple flasks
SEQ ID NO:6
SEQ ID NO:7
Step 5
Pooling samples
Add primer B, denature, anneal
to Fig. 9B
from Fig. 9A
SEQ ID NO:7
Product 5
SEQ ID NO:6
SEQ ID NO:7
Step 6
Polymerase with 3'-5' proofreading
SEQ ID NO:6
Product 6
Clean
Add PrA and PrB, denature, anneal
SEQ ID NO:7
SEQ ID NO:6
SEQ ID NO:7
Step 7
SEQ ID NO:6
Product 7
SEQ ID NO:7
SEQ ID NO:6
Step 8
PCR
SEQ ID NO:6
Product 8
SEQ ID NO:6
Step 9
Library preparation
SEQ ID NO:7
Product 9
Library
Sequencing

Fig. 9

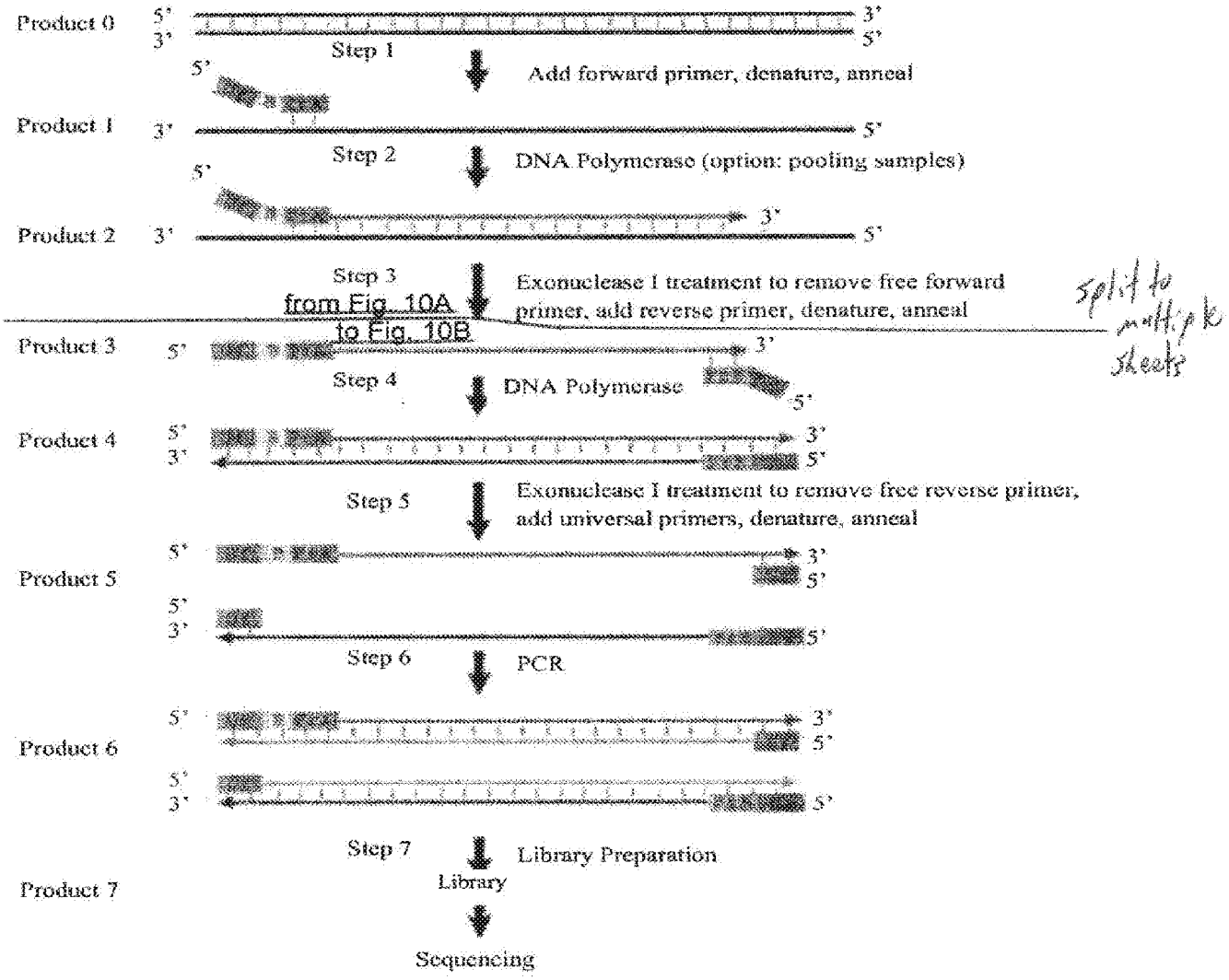

Product 0
5'
3'
3'
5'
Step 1
Add forward primer, denature, anneal
5'
Product 1
3'
5'
Step 2
DNA Polymerase (option: pooling samples)
5'
Product 2
3'
3'
5'
Step 3
Exonuclease I treatment to remove free forward primer, add reverse primer, denature, anneal
from Fig. 10A
to Fig. 10B
Split to multiple sheets
Product 3
5'
3'
Step 4
DNA Polymerase
5'
Product 4
5'
3'
3'
5'
Step 5
Exonuclease I treatment to remove free reverse primer, add universal primers, denature, anneal
5'
3'
Product 5
5'
3'
5'
Step 6
PCR
5'
3'
Product 6
5'
3'
5'
Step 7
Library Preparation
Library
Product 7
Sequencing

Fig. 10

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension - 3 months with $0 paid	2253	1	700	700
Miscellaneous:				
			Total in USD ($)	**700**

Electronic Acknowledgement Receipt

EFS ID:	34041181
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	17-OCT-2018
Filing Date:	07-MAR-2016
Time Stamp:	18:09:48
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$700
RAM confirmation Number	101818INTEFSW00005109033125
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.293

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		181017_81503-AA_Amendment_ADR.pdf	753389 c8814d3af3a593f16ac36b1e590f064c7f44ba3f	yes	19

Multipart Description/PDF files in .zip description

Document Description	Start	End
Amendment/Req. Reconsideration-After Non-Final Reject	1	1
Drawings-only black and white line drawings	2	2
Specification	3	3
Claims	4	11
Applicant Arguments/Remarks Made in an Amendment	12	18
Amendment/Req. Reconsideration-After Non-Final Reject	19	19

Warnings:

Information:

2	Drawings-only black and white line drawings	181017_81503-AA_Exhibit_A_ADR.pdf	2863269 01d2dcc91f80965e150cc7deb98e2c1be4c7a5fa	no	8

Warnings:

Information:

3	Drawings-only black and white line drawings	181017_81503-AA_Exhibit_B_ADR.pdf	3275795 cf96baf05dcab7bc5c936688d3a67752ee1d6b64	no	4

Warnings:

Information:

4	Fee Worksheet (SB06)	fee-info.pdf	30910 6e2d14a0da282874ae0a256cc57fbe6de7dbe414	no	2

Warnings:	
Information:	
Total Files Size (in bytes):	6923363

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

REMARKS

Claims 68-87 are pending in the subject application with claim 76 withdrawn by the Examiner. By this Amendment, applicants have hereinabove amended claims 68-74 and 76-87 to correct the number of claim dependencies and for the purposes of clarity.

Applicants maintain that the amendment presented hereinabove raise no issue of new matter and respectfully request that this Amendment be entered. Accordingly, upon entry of this Amendment, claims 68-75 and 77-87 are pending and under consideration in the subject application with claim 76 withdrawn from consideration by the Examiner.

Objections to the Drawings

In the April 17, 2018 Office Action, the Examiner objected to Figures 7, 9 and 10 for allegedly containing letters that were difficult to recognize.

Applicants' reply

In response, applicants attach hereto as **Exhibit A** EIGHT (8) replacement sheets of drawings containing Figures 7, 9, and 10. Applicants submit that the letters in replacement Figures 7, 9 and 10 are clear. Additionally, sequences having more than 10 nucleotides are identified by their respective SEQ ID NO in Figures 8 and 9. Additionally, applicants attach as **Exhibit B**, FOUR (4) sheets of drawings containing previous Figures 7-10, marked to show the amendments.

Applicants have also amended the description of Figures 7, 9 and 10 in the subject application to reflect the multiple views of the amended figures.

Applicants maintain that amendments to the drawings and the

specification raise no issue of new matter.

Accordingly, applicants respectfully request that the objections to the drawings be withdrawn.

Objections to the Specification

In the April 17, 2018 Office Action, the Examiner required applicants to update the priority status of the subject application. The Examiner further objected to the specification because the BRIEF DESCRIPTION OF THE FIGURES only described Figure 5, but there are Figures 5A and 5B. Additionally, neither the descriptions of Figures 8 and 9 nor the Figures themselves identified the SEQ ID NO for nucleotides in Figures 8 and 9 that had more than 10 nucleotides.

Applicants' reply

In response, applicants have updated the priority information for the subject application. Additionally, applicants have amended the description of Figure 5 in the BRIEF DESCRIPTION OF THE FIGURES of the subject application do describe Figures 5A and 5B. As discussed above, Figures 8 and 9 have been amended to identify the SEQ ID NO for oligonucleotides therein.

Applicants maintain that the amendments to the specification raise no issue of new matter.

Accordingly, applicants respectfully request that the objections to the specification be withdrawn.

Objections to the Claims

In the April 17, 2018 Office Action, the Examiner objected to claims 68, 81, and 85. The Examiner's rationale and suggested claim amendments may be found on page 3 of the April 17, 2018 Office Action.

Applicants' reply

In response, without conceding correctness of the Examiner's position, and solely for the purpose of advancing prosecution, applicants have amended claims 68, 81, and 85 without prejudice or disclaimer. The amendments to these claims are consistent with the Examiner's suggested claim amendments on page 3 of the April 17, 2018 Office Action.

Accordingly, applicants respectfully request that the claim objections be withdrawn.

Rejection of claims under 35 U.S.C. § 112, Second Paragraph

In the April 17, 2018 Office Action the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 112, second paragraph, as allegedly indefinite for failing to particularly point out and distinctly claim the subject matter with applicants regard as the invention. The Examiner's rationale is set forth on pages 3 and 4 of the April 17, 2018 Office Action.

Applicants' reply

As an initial matter, without conceding correctness of the Examiner's position, and solely for the purpose of advancing prosecution, applicants have amended claims 68, 69, and 77. Applicants submit that the amendments to claims 68, 69 and 77 render moot the Examiner's rejection of claims 69, 70, 71, 72, 74, and 77.

Further, applicants respectfully traverse the Examiner's rejections as they may apply to claims 68 and 75 as amended. Applicants submit that the term "corresponding to" in claim 68 is not unclear. Rather, the Specification clearly defines the meaning of the term "corresponding to." For example, with regard to corresponding tag associated sequence reads, the Specification states that:

> [a]s used herein, a subsequence of a tag associated sequence read is 'corresponding to' a tag when the subsequence is identical to the nucleotide sequence of the tag.

(Specification at pg. 26, lines 15-16). With regard to a species of nucleic acid molecule, the Specification states that:

> [a]s used herein, a subsequence of a tag associated sequence read is 'corresponding to' a species of nucleic acid molecule when the subsequence is substantially identical to or substantially complementary to at least about 10, 12, 14, 16, 18, 20, or more nucleotides of the sequence of the species of nucleic acid molecule. In an embodiments the subsequence is identical to or fully complementary to at least about 10, 12 14, 16, 18, 20, or more nucleotides of the sequence of the species of nucleic acid molecule.

(Specification at pg. 26, lines 17-22).

Furthermore, applicants submit that the Specification clearly discloses embodiments of how copy number information can be obtained from the genomic nucleic acids based on claim 68, step f) or the mRNA transcripts claim 25, step e). Specifically, the Specification states that:

> There are two separate methods of counting, and each can be combined for greater accuracy and determination of confidence.
>
> The first counting method is called "maximum tagged nucleic acid molecule number." The number of different tagged nucleic acid molecules at a given location cannot exceed the true (absolute) copy number. Therefore, for a given region comprising multiple locations, the maximum copy number of the region is not less than the maximum number of different tagged nucleic acid molecules mapped to any location in that region. The maximum number of different tagged nucleic acid molecules centered about a moving window of a fixed number of locations can be taken as the measurement of true copy number of the window. This will provide at worst an underestimate of the maximum true copy

> number for that window, and never an overestimate. The set of maximum number of different tagged nucleic acid molecules is one type of copy number profile.
>
> The counting method described above is most accurate when the processing efficiency is excellent and the number of molecules mapped to each location is less than the number of tags. Under such conditions virtually each molecule in the sample is tagged and counted, and the true count can be derived. But there is a second method.
>
> The second counting method is called "total different tagged nucleic acid molecules," and is useful, for example, when tagging efficiency is low. The total number of different tagged nucleic acid molecules within a region of a fixed number of locations will be, within a statistical counting error, a monotonic function of true copy number, regardless of the efficiency of tagging. Low efficiency of tagging merely increases the statistical error. The relative copy number of a region can be estimated by comparing the total number of different tagged nucleic acid molecules mapped to the region to, for example, the mean value of tagged nucleic acid molecules mapped to other regions of the genome which have a comparable number of locations.

Applicants submit that the pending claims comply with 35 U.S.C. § 112, second paragraph and respectfully request that this rejection be withdrawn.

Rejection of claims under 35 U.S.C. § 101

In the April 17, 2018 Office Action the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 101, as allegedly not directed to patent eligible subject matter. The Examiner's rationale is set forth on pages 5-7 of the April 17, 2018 Office Action.

Applicants' reply

In response, applicants respectfully traverse the Examiner's rejection.

Applicants submit that the pending claims are directed to a "method"

for obtaining sequencing information from nucleic acids comprising specific physical steps, rather than to an abstract idea.

Applicants remind the Examiner that the MPEP advises "that examiners should consider the claim as a whole when performing the Step 2A analysis." (MPEP §2106.04, paragraph II, titled "Eligibility Step 2A, Whether a Claim is Directed to a Judicial Exception"). The MPEP further advises that "[s]ome claims are not directed to an abstract idea because they do not recite anything similar to a judicially-identified abstract idea, although it may be apparent that at some level they are based on or involve an abstract idea." (MPEP §2106.04(a)(1), paragraph I).

Specifically, the MPEP provides "[n]on-limiting hypothetical examples of claims that do not set forth or describe an abstract idea," among which are included "[a] method for sequencing BRCA1 gene sequences comprising: amplifying by polymerization chain reaction technique all or part of a BRCA1 gene from a tissue sample from a human subject using a set of primers to produce amplified nucleic acids; and sequencing the amplified nucleic acids." (MPEP §2106.04(a)(1), paragraph I).

The claims pending in the subject application recite a method comprising amplifying nucleic acids to produce amplified nucleic acids and sequencing the amplified nucleic acids. In fact, in comparison to the M.P.E.P. provided example of a claim that is patent eligible, the pending claims further require the physical steps of "tagging" segments of genomic nucleic acids with nucleic acid tags, thereby generating unique tagged nucleic acid molecules prior to PCR and mapping the genomic nucleic acids.

Applicants submit that the pending claims comply with 35 U.S.C. § 101 and respectfully request that this rejection be withdrawn.

Non-statutory Double Patenting

In response, applicants respectfully request that the rejection under non-statutory double patenting over claims 1-20 of U.S. Patent No. 9,404,156 B2 be held in abeyance until allowable claims have been identified.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the $700.00 fee for a three-month extension of time for a small entity, is deemed necessary in connection with the filing of this Amendment. However, if any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

2018-10-17
Aditya Ramanathan Date

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 11/30/2018

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
11/30/2018	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.304

Office Action Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☑ Responsive to communication(s) filed on 10/17/2018.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☑ This action is **FINAL.** 2b) ☐ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5) ☑ Claim(s) 68-87 is/are pending in the application.

5a) Of the above claim(s) 76 is/are withdrawn from consideration.

6) ☐ Claim(s) _____ is/are allowed.

7) ☑ Claim(s) 68-75 and 77-87 is/are rejected.

8) ☐ Claim(s) _____ is/are objected to.

9) ☐ Claim(s) _____ are subject to restriction and/or election requirement

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to **PPHfeedback@uspto.gov.**

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☑ The drawing(s) filed on 10/17/2018 is/are: a)☐ accepted or b)☑ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)

2) ☐ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date _____.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date _____.

4) ☐ Other: _____.

GUARDANT HEALTH EX.1009.305

DETAILED ACTION

Response to Amendment

1. Applicant's response to the office action filed on October 17, 2018 has been entered. The claims pending in this application are claims 68-87 wherein claim 76 has been withdrawn due to the restriction requirement mailed on January 18, 2018. Rejection and/or objection not reiterated from the previous office action are hereby withdrawn in view of applicant's amendment filed on October 17, 2018. Claims 68-75 and 77-87 will be examined.

Drawings

2. Applicant submitted new Figures 7A, 7B, 8, 9A, 9B, 10A and 10B on October 17, 2018 to replace Figures 7 to 10. However, since some letters in Figures 7A, 7B, 8, 9A, 9B, 10A and 10B are difficult to be recognized, applicant is required to furnish a drawing under 37 CFR 1.81(c). No new matter may be introduced in the required drawing. Each drawing sheet submitted after the filing date of an application must be labeled in the top margin as either "Replacement Sheet" or "New Sheet" pursuant to 37 CFR 1.121(d).

Claim Objections

3. Claim 68 is objected to because of the following informality: "thereby obtaining genomic copy number information from the genomic nucleic acids" in step f) should be "and obtaining genomic copy number information from the genomic nucleic acids".

4. Claim 69 is objected to because of the following informality: "within a region of the genome" should be deleted.

5. Claim 77 is objected to because of the following informality: "unique tagged nucleic acid" in last line should be "the unique tagged nucleic acid".

6. Claim 81 is objected to because of the following informality: "product of step (c)" in last line should be "of step (c)".

7. Claim 85 or 87 is objected to because of the following informality: "subject to" should be "subjected to". Note that applicant has not addressed this issue.

Appropriate correction is required.

Claim Rejections - 35 USC § 112

8. The following is a quotation of 35 U.S.C. 112(b):

(b) CONCLUSION.—The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the inventor or a joint inventor regards as the invention.

The following is a quotation of 35 U.S.C. 112 (pre-AIA), second paragraph:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

9. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 112(b) or 35 U.S.C. 112 (pre-AIA), second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which the inventor or a joint inventor, or for pre-AIA the applicant regards as the invention.

10. Claim 68 is rejected as vague and indefinite. Since step f) only requires counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome, a count for the same location on the genome can be obtained. However, a count for each location on the genome and genomic copy number

information from the genomic nucleic acids (eg., different genomic nucleic acids) cannot be obtained only based on step f). Please clarify.

11. Claim 70 or 71 or 72 or 74 recites the limitation "the counts obtained in step (f)" in the claim. There is insufficient antecedent basis for this limitation in the claim because there is no word "counts" in step f) of claim 68. Please clarify. Note that applicant has not response this issue.

12. Claim 75 is rejected as vague and indefinite because it is unclear how mRNA copy number information can obtained from the mRNA transcripts only based on step e) because the claim does not indicate what is the relationship between mRNA copy number information and the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library. Since step e) only requires counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, only a count for the same location on the cDNA library can be obtained. Please clarify.

Claim Rejections - 35 USC § 101

13. 35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

14. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 101 because

The claimed invention is directed to a judicial exception (i.e., a law of nature, a natural phenomenon, or an abstract idea) without significantly more. Claims 68-74 and 77-87 are directed to a process of obtaining genomic copy number information from genomic nucleic acids while claim 75 is directed to a process of obtaining mRNA copy number information from

mRNA transcripts. The claims do not include additional elements that are sufficient to amount to significantly more than the judicial exception because:

The Supreme Court has identified a number of considerations for determining whether a claim with additional elements amounts to significantly more than the judicial exception itself. The following are examples of these considerations, which are not intended to be exclusive or limiting. Limitations that may be enough to qualify as "significantly more" when recited in a claim with a judicial exception include:

• Applying the judicial exception with, or by use of, a particular machine;

• Effecting a transformation or reduction of a particular article to a different state or thing;

• Adding a specific limitation other than what is well-understood, routine and conventional in the field, or adding unconventional steps that confine the claim to a particular useful application; or

• Other meaningful limitations beyond generally linking the use of the judicial exception to a particular technological environment.

Limitations that were found not to be enough to qualify as "significantly more" when recited in a claim with a judicial exception include:

• Adding the words "apply it" (or an equivalent) with the judicial exception, or mere instructions to implement an abstract idea on a computer;

• Simply appending well-understood, routine and conventional activities previously known to the industry, specified at a high level of generality, to the judicial exception;

• Adding insignificant extrasolution activity to the judicial exception; or

• Generally linking the use of the judicial exception to a particular technological environment or field of use.

A two-step procedure is used to determine if claimed subject matter is a judicial exception. First it is determined whether the claimed subject matter is directed to a law of nature, a natural phenomenon, or an abstract idea. The processes of claims 68-74 and 77-87 are directed to a process of obtaining genomic copy number information from genomic nucleic acids wherein

steps e) and f) in independent claim 68 and dependent claims 69-72, 74, and 82 are considered as mental steps. The process of claim 75 is directed to a process of obtaining mRNA copy number information from mRNA transcripts wherein steps d) and e) are considered as mental steps. **Note that steps in a claim that recite the manipulation of information through a series of mental steps would be considered a single abstract idea for purposes of analysis** (see MPEP 2106.04).

In the second step it is determined whether the claimed subject matter includes elements that amount to significantly more than the judicial exception. The claims have additional limitations that constitute conventional steps appended to the abstract ideas. In independent claim 68, the steps a) to d) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see claims 1-21 of US Patent No. 9,670,536 B2 with a priority date of September 21, 2010) and pages 119 and 120, online methods and Figure 1 from Hiatt *et al.,* Nature Methods, 7, 119-122, published online on January 17, 2010) while, in independent claim 75, the steps a) to c) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see Figures 1a, 1c and 1d in page 529 and online methods from Plessy *et al.,* Nature Methods, 7, 528-534, published online on June 13, 2010). The conventional steps do not comprise an inventive concept that transforms the claimed abstract ideas into a patent-eligible application of the abstract ideas. The claimed subject matters of claims 68-75 and 77-87 are processes directed to abstract ideas. When recited at this high level of generality, there is no meaningful limitation, such as a particular or unconventional machine or a transformation of a particular article, in this step that distinguishes it from well-understood, routine, and conventional data gathering activity engaged in by scientists prior to applicant's invention, and at

the time the application was filed, *e.g.*, the routine and conventional techniques for performing steps a) to d) of claim 68 or steps a) to c) of claim 75. Further, it is well established that the mere physical or tangible nature of additional elements such as steps a) to d) of claim 68 or steps a) to c) of claim 75 do not automatically confer eligibility on a claim directed to an abstract idea (*see, e.g.*, Alice Corp. *v.* CLS Bank Int'l, 134 S. Ct. 2347, 2358-59 (2014)). Therefore, the claimed subject matters of claims 68-75 and 77-87 are without limitations that indicate that the claims are no different from the process claims directed to abstract ideas and are patent-ineligible.

Response to Arguments

In page 16, fifth paragraph bridging to page 17, last paragraph of applicant' remarks, applicant argues that "[A]pplicants submit that the pending claims are directed to a 'method' for obtaining sequencing information from nucleic acids comprising specific physical steps, rather than to an abstract idea. Applicants remind the Examiner that the MPEP advises 'that examiners should consider the claim as a whole when performing the Step 2A analysis.' (MPEP §2106.04, paragraph II, titled 'Eligibility Step 2A, Whether a Claim is Directed to a Judicial Exception') . The MPEP further advises that '(s]ome claims are not directed to an abstract idea because they do not recite anything similar to a judicially-identified abstract idea, although it may be apparent that at some level they are based on or involve an abstract idea.' (MPEP §2106.04(a)(1), paragraph I). Specifically, the MPEP provides '[n]on-limiting hypothetical examples of claims that do not set forth or describe an abstract idea,' among which are included '[a] method for sequencing BRCAl gene sequences comprising: amplifying by polymerization chain reaction technique all or part of a BRCAl gene from a tissue sample from a human subject using a set of primers to produce amplified nucleic acids; and sequencing the amplified nucleic acids.' (MPEP §2106.04(a)(1), paragraph I).The claims pending in the subject application recite a method

comprising amplifying nucleic acids to produce amplified nucleic acids and sequencing the amplified nucleic acids. In fact, in comparison to the M.P.E.P. provided example of a claim that is patent eligible, the pending claims further require the physical steps of 'tagging' segments of genomic nucleic acids with nucleic acid tags, thereby generating unique tagged nucleic acid molecules prior to PCR and mapping the genomic nucleic acids".

The above argument has been fully considered but it is not persuasive toward the withdrawal of the rejection. Although applicant argues that "the MPEP provides '[n]on-limiting hypothetical examples of claims that do not set forth or describe an abstract idea,' among which are included '[a] method for sequencing BRCAl gene sequences comprising: amplifying by polymerization chain reaction technique all or part of a BRCAl gene from a tissue sample from a human subject using a set of primers to produce amplified nucleic acids; and sequencing the amplified nucleic acids.' (MPEP §2106.04(a)(1), paragraph I)", the office action have clearly indicated that "[I]n independent claim 68, the steps a) to d) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see claims 1-21 of US Patent No. 9,670,536 B2 with a priority date of September 21, 2010) and pages 119 and 120, online methods and Figure 1 from Hiatt *et al.,* Nature Methods, 7, 119-122, published online on January 17, 2010) while, in independent claim 75, the steps a) to c) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see Figures 1a, 1c and 1d in page 529 and online methods from Plessy *et al.,* Nature Methods, 7, 528-534, published online on June 13, 2010)". Since steps e) and f) of claim 68 or steps d) and e) of claim 75 are considered as mental steps, the conventional steps a) to d) of claim 68 or steps a) to c) of claim 75 do not comprise an inventive concept that transforms the claimed abstract ideas into a patent-eligible application of the

abstract ideas. The claimed subject matters of claims 68-75 and 77-87 are processes directed to abstract ideas. **Note that steps in a claim that recite the manipulation of information through a series of mental steps would be considered a single abstract idea for purposes of analysis** (see MPEP 2106.04).

Double Patenting

15. The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. A nonstatutory double patenting rejection is appropriate where the conflicting claims are not identical, but at least one examined application claim is not patentably distinct from the reference claim(s) because the examined application claim is either anticipated by, or would have been obvious over, the reference claim(s). See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) or 1.321(d) may be used to overcome an actual or provisional rejection based on nonstatutory double patenting provided the reference application or patent either is shown to be commonly owned with the examined application, or claims an invention made as a result of activities undertaken within the scope of a joint research agreement. See MPEP § 717.02 for applications subject to examination under the first inventor to file provisions of the AIA as explained in MPEP § 2159. See MPEP

§§ 706.02(l)(1) - 706.02(l)(3) for applications not subject to examination under the first inventor to file provisions of the AIA. A terminal disclaimer must be signed in compliance with 37 CFR 1.321(b).

The USPTO Internet website contains terminal disclaimer forms which may be used. Please visit www.uspto.gov/patent/patents-forms. The filing date of the application in which the form is filed determines what form (e.g., PTO/SB/25, PTO/SB/26, PTO/AIA/25, or PTO/AIA/26) should be used. A web-based eTerminal Disclaimer may be filled out completely online using web-screens. An eTerminal Disclaimer that meets all requirements is auto-processed and approved immediately upon submission. For more information about eTerminal Disclaimers, refer to www.uspto.gov/patents/process/file/efs/guidance/eTD-info-I.jsp.

16. Claims 68-75 and 77-87 are rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 1-20 of U.S. Patent No. 9,404,156 B2. Although the conflicting claims are not identical, they are not patentably distinct from each other because the examined claims in this instant application are either anticipated by, or would have been obvious over, the reference claims. See *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); and, *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969). Although claims 68-75 and 77-87 in this instant application are not identical to claims 1-20 of U.S. Patent No. 9,404,156 B2, claims 1-20 of U.S. Patent No. 9,404,156 B2 are directed to the same subject matter and fall entirely within the scope of claims 68-75 and 77-87 in this instant application. In other words, claims 68-75 and 77-87 in this instant application are anticipated by claims 1-20 of U.S. Patent No. 9,404,156 B2.

Response to Arguments

In page 18, first paragraph of applicant' remarks, applicant argues that "applicants respectfully request that the rejection under non-statutory double patenting over claims 1-20 of U.S. Patent No. 9,404,156 B2 be held in abeyance until allowable claims have been identified".

The above argument has been fully considered but it is not persuasive toward the withdrawal of the rejection because applicant has not filed a terminal disclaimer yet.

Conclusion

19. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

20. No claim is allowed.

21. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30

(November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993)(See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/FRANK W LU/
Primary Examiner, Art Unit 1634
November 28, 2018

Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	Art Unit
	FRANK W LU	1634

CPC - Searched*		
Symbol	Date	Examiner
Update	11/27/2018	/FL/

CPC Combination Sets - Searched*		
Symbol	Date	Examiner
Update	11/27/2018	/FL/

US Classification - Searched*			
Class	Subclass	Date	Examiner
Update		11/27/2018	/FL/

* See search history printout included with this form or the SEARCH NOTES box below to determine the scope of the search.

Search Notes		
Search Notes	Date	Examiner
Update	11/27/2018	/FL/

Interference Search			
US Class/CPC Symbol	US Subclass/CPC Group	Date	Examiner

/FRANK LU/ Primary Examiner,Art Unit 1634	November 27, 2018

GUARDANT HEALTH EX.1009.317

Substitute Form PTO-1449 (Modified) U.S. Department of Commerce Patent and Trademark Office	Attorney Docket No. 81503-AA/GJG/ADR	Application No. 15/063,278
Information Disclosure Statement by Applicant (Use several sheets if necessary)	First Named Inventor James HICKS	Confirmation Number 4038
(37 CFR §1.98(b))	Filing Date March 7, 2016	Group Art Unit 1634

U.S. Patent Documents

Examiner Initial	Cite ID[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM/DD/YYYY	Inventor(s) or Applicant(s)
	3.	2008-0261204	10/23/2008	LEXOW
	4.	2007-0020640	01/25/2007	MCCLOSKEY et al.

FOREIGN PATENT DOCUMENTS

Examiner Initial	Cite ID[1]	Foreign Patent Document Country Code[3] Number[4] Kind Code[5] (if known)	Publication Date MM/DD/YYYY	Inventor(s)	Country or Patent Office[3]	T[6]
	5.	DE 10 2008 025 656 A1	12/03/2009	Bjorn ROTTER	DE	

NON PATENT LITERATURE DOCUMENTS

Examiner Initial	Cite ID[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.
	1.	Communication of a notice of opposition, issued by the European Patent Office on February 1, 2019, concerning European Patent No. EP2630263
	2.	Notice of opposition to a European Patent, filed January 25, 2019, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263
	6.	Chiang et al. (2008) "High-resolution Mapping of Copy-number Alterations with Massively Parallel Sequencing", Nature Methods 6(1):99-103
	7.	Hug and Schuler (2003) "Measurement of the Number of Molecules of a Single mRNA Species in a Complex mRNA Preparation", J. theor. Biol. 221:615-624
	8.	Meyerson et al. (2010) "Advances in Understanding Cancer Genomes Through Second-Generation Sequencing", Nature Reviews 11:685-696

Examiner Signature	Date Considered

***EXAMINER:** Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

German Offenlegungsschrift

Title: **Method for the quantitative analysis of nucleic acids, markers therefor and their use**

Abstract: The invention relates to a method for the quantitative determination of nucleic acids in a sample, especially for the quantitative determination of gene transcripts, for example mRNA, cDNA, microRNA, non-coding RNA, and to the provision of markers for conduction of the analysis methods.

U.S. Serial No.: 15/063,278
Applicant(s): James Hicks et al.
Filed: March 7, 2016
Exhibit 3

GUARDANT HEALTH EX.1009.319

Description:

[0001] The invention relates to a method for the quantitative determination of nucleic acids in a sample, especially for the quantitative determination of gene transcripts, genomic DNA or genomic nucleic acid sections, for example DNA, RNA, mRNA, cDNA, microRNA, non-coding RNA, aptamer-DNA and -RNA, and to the provision of markers for conduction of the analysis methods.

[0002] The quantification of nucleic acids has great scientific and commercial significance in all fields of life sciences. Firstly, it is important to know which gene products (for example protein-encoding and non-coding RNA) occur how frequently (gene expression studies), and secondly how transcription is regulated, for example by epigenetic properties of the genomic DNA. In addition, the determination of the frequency of copies of gene sections for karyotyping is an important diagnosis option for diseases such as cancer, for example.

[0003] This need has led to the development of various methods which are generally either based on the unspecific measurement of the total amount of all or particular nucleic acids, for example by photometric methods (total RNA or DNA), or are intended to individually quantify a multitude of different nucleic acid molecules in a particular sequence. The methods used for the purpose can basically be classified into 2 groups: the first group is that of methods which can quantify already known or otherwise characterized sequences. They are generally based on binding of the nucleic acids to be quantified to known or otherwise characterized sequences bound to a solid surface. Examples are all forms of the Southern blot and

GUARDANT HEALTH EX.1009.320

Northern blot, and what are called DNA or RNA micro-arrays (chips).

[0004] The second group is that of methods which are based on the quantification of the nucleic acids by sequencing of a maximum number of individual nucleic acid molecules and counting of the sequenced molecules in the mixture. Since it is generally the case that more nucleic acid molecules are present in the mixture than are sequenced, the relative frequency of a nucleic acid of a particular sequence in the mixture is thus obtained.

[0005] The invention presented here improves the reliability and accuracy of the quantification of nucleic acids by virtually all methods that are based on sequencing and counting. These include both what are called "tag"-based methods, wherein the tag is a fragment which is representative of the nucleic acid to a maximum degree, on the basis of which the nucleic acid can be identified, and methods in which the number of complete DNA and RNA molecules, for example those of viruses, is determined.

[0006] Because of the development of techniques for simultaneous sequencing of hundreds of thousands to millions of nucleic acid molecules, sequence-based quantitative methods are finding ever broader use and are replacing qualitative, array-based methods in many areas. Therefore, sequence-based quantification will gain ever higher significance in the future. A list of the most important sequence-based quantification methods for nucleic acids can be found in S.M. Wang (2007) Understanding SAGE data, Trends in Genetics 23: 42-50. Specifically, these include:

- "Serial analysis of gene expression" (SAGE, V.E. Velculescu et al., Serial analysis of gene

expression, Science 270 (1995), pp. 484-487, US Patent 5,695,937): SAGE facilitates the global quantitative characterization of a transcriptome by using a type II restriction enzyme (BsmFI) as a "tagging enzyme", in order to cut a fragment of a length 14 Bp, called the "tag", out of a cDNA beyond the farthest restriction site toward the 3' end for a frequent-cutting restriction enzyme (usually beyond the NIaIII restriction site CATG). In the case of SAGE, 2 tags in each case are ligated together head-to-head to give what are called ditags, which are then connected ("concatemerized") to form longer chains ("concatemers"). These concatemers are then cloned and sequenced. The tags represent the starting mRNA molecules which have generated the cDNA. Therefore, the amount of tags sequenced constitutes a measure of the relative frequency with which the mRNA was represented in the mRNA mixture. Nowadays, concatemer formation and cloning can be dispensed with because the ditags can be sequenced after a PCR directly with highly parallel sequencing techniques, for instance the above-described methods (e.g. Nielsen KL, Høgh AL, Emmersen J., Nucleic Acids Res. 2006; 34(19): e133. DeepSAGE – digital transcriptomics with high sensitivity, simple experimental protocol and multiplexing of samples).

- "LongSAGE" (S. Saha et al., Using the transcriptome to annotate the genome, *Nat. Biotechnol.* 20 (2002), pp. 508-512) was developed in order to increase the specificity of the SAGE tags having a length of only 14 Bp by using another type II restriction enzyme (MmeI) to generate tags of length 21 Bp. This improves the assignment to genomic or EST sequences.

GUARDANT HEALTH EX.1009.322

Quantification with the SAGE method and especially with LongSAGE is problematic; for example, it is often necessary to rectify replication-related distortions in the quantification by biostatistical analysis methods (e.g. Emmersen, J. et al., BMC Bioinformatics 2007, 8:92). However, these methods are based on statistical assumptions and can therefore only approximately estimate the actual relative frequency of the tags in a sample.

- "SuperSAGE" (H. Matsumura et al., Gene expression analysis of plant host-pathogen interactions by SuperSAGE, *Proc. Natl. Acad. Sci. U. S. A.* 100 (2003), pp. 15718-15723) provides tags of length 25-27 Bp through use of the type III restriction enzyme EcoP15I as tagging enzymes, which have further advantages as well as the further improvement in assignment to other sequences.

- "Cap analysis gene expression" (CAGE, T. Shiraki et al., Cap analysis gene expression for high-throughput analysis of transcriptional starting point and identification of promoter usage, *Proc. Natl. Acad. Sci. U. S. A.* 100 (2003), pp. 15776-15781) was developed in order to identify transcript initiation sequences and promoters. In the method, fragments of length 21 Bp which directly adjoin the 5'-CAP sequence of an mRNA are isolated and sequenced.

- "Gene identification signature" (GIS, C.L. Wei et al., 5' Long serial analysis of gene expression (LongSAGE) and 3' LongSAGE for transcriptome characterization and genome annotation, *Proc. Natl. Acad. Sci. U. S. A.* 101 (2004), pp. 11701-11706) serves for simultaneous determination of the 5' and 3' ends of mRNAs. GIS tags have a length of 20 Bp and can be used to identify the ends of a gene in the genome.

GUARDANT HEALTH EX.1009.323

- "Digital karyotyping" DK; (T.L. Wang et al., Digital karyotyping, *Proc. Natl. Acad. Sci. U. S. A.* 99 (2002), pp. 16156-16161) adapts the LongSAGE protocol in order to obtain 21 Bp tags from genomic DNA. It is thus possible to exactly determine the pathological replication or loss of particular chromosome sections and insertions and foreign DNA in cancer cells, for example (J.T. Park et al., Notch3 gene amplification in ovarian cancer, *Cancer Res.* 66 (2006), pp. 6312-6318).
- "Methylation-specific digital karyotyping" (MSDK) (Min Hu et al., Methylation-specific digital karyotyping Nature Protocols 1, 1621-1636 (2006)) applies the principle of digital karyotyping for the quantitative determination of different methylation states in two samples.
- "Paired-end ditag" (C.L. Wei et al., A global map of p53 transcription-factor binding sites in the human genome, *Cell.* 124 (2006), pp. 207-219) has the aim of identifying protein-binding sequences in the genome. This uses the principle of GIS to isolate and to quantify tags from both ends of a DNA fragment which are bound to a particular protein (e.g. a transcription factor), and said protein is precipitated specifically from the protein-DNA mixture together with the bound DNA by immunoprecipitation. Paired-end ditag was used in order to identify p53 binding sites in the human genome.
- The aim of metagenomic analyses is to determine the composition of a complex mixture of organisms. This uses the same methods as digital karyotyping. Krause et al., Phylogenetic classification of short environmental DNA fragments, Nucleic Acids Res. 2008 April; 36(7): 2230-2239.

[0007] In all the methods described, the amount of nucleic acids available in a sample is usually insufficient for all the subsequent analyses. For that reason, replication of the material is almost always necessary prior to the analysis.

[0008] For example, highly parallel sequencing methods (e.g. the "454" picoliter method from Roche Diagnostics, Germany, or the Solexa pyrosequencing method from Illumina, Inc., San Diego, USA) require 5-3000 ng of DNA. In addition, both methods require specific sequences (adapters) at both ends of the nucleic acid to be sequenced, which serve as primer binding site. In order to assure the presence of these specific sequences in all molecules to be sequenced, the polymerase chain reaction (PCR) is utilized, using oligonucleotides complementary to the adapter sequences as primers. For cloning of the fragments too, the prior amplification thereof is often necessary.

[0009] However, amplification, for example by means of PCR, harbors the risk that particular nucleic acids will be replicated preferentially. For instance, short nucleic acids are amplified preferentially, but the individual base sequence can also affect the efficiency of amplification. As a result, the relative frequency of a nucleic acid in any nucleic acid population may differ greatly before and after the amplification.

[0010] Frequently, moreover, for instance in the quantification of expressed sequenced tags (ESTs), the quantification of nucleic acid compositions is also carried out without prior PCR amplification, as for example after cloning of the nucleic acids. In this case as well, there may be a distortion of the original composition of the nucleic acid population, owing to differences in fitness of individual bacteria and owing

GUARDANT HEALTH EX.1009.325

to effects of the nucleic acids on the growth of the transformed cells.

[0011] It was an object of the present invention, accordingly, to provide a simple method in which the distortions in the nucleic acid compositions that occur during the replication of the nucleic acids in a sample that are to be quantified can be more effectively recognized and corrected in the context of a quantitative evaluation.

[0012] The object is achieved by providing all nucleic acids in a nucleic acid mixture, irrespective of whether they are double-stranded or single-stranded, with a "random tag" (RAT) prior to the amplification, this random tag originating from a mixture of synthetic oligonucleotides, and the mixture comprising sufficient RATs with different sequences to ensure that the resulting random tag-nucleic acid (RAT-NS) combinations are virtually unique. The techniques by which the nucleic acids can be provided with a RAT prior to amplification may be very different. After sequencing, it is possible to ascertain the RAT-nucleic acid combinations, and a data set is produced with the individual sequence information of the RAT-NS. RAT-nucleic acid combinations which occur more than once or at least more frequently than statistically probable are therefore recognizable as copies. Elimination of all the copies from the data set produces the original composition of the nucleic acid mixture prior to the amplification.

[0013] The subject of the present invention, accordingly, is a method for the quantitative determination of nucleic acids (nucleic acid population) in a sample, wherein

a) nucleic acid constructs comprising

GUARDANT HEALTH EX.1009.326

- at least one characterizing polynucleotide domain (TAG) of a nucleic acid which is present in a sample and which is to be quantitatively determined, and
- at least one artificial polynucleotide domain with random sequence ("random tag" or RAT for short), and
- at least one primer binding site or polymerase binding site (PB), where one characterizing polynucleotide domain (TAG) and at least one polynucleotide domain with random sequence (RAT) form a characteristic domain combination (TAG-RAT combination) in each case, which is flanked by at least one polymerase binding site or primer binding site (PB), are amplified in the presence of a polymerase, and

b) after the amplification, at least the sequence of the polynucleotide domains with random sequence (RAT) and at least a part of the characterizing polynucleotide domain (TAG) are determined, it being possible, for the quantitative determination of the nucleic acid present in the sample, for combinations of identical sequence, or for combinations of identical sequence and combinations of similar sequence, of a characterizing polynucleotide domain (TAG) and of a polynucleotide domain with random sequence (RAT) to be identified as copies of a characterizing polynucleotide domain (TAG) of the nucleic acid originally present in the sample.

[0014] By "nucleic acid" in the sense of the present invention is meant the nucleic acids which are present in a sample and which are intended for quantitative determination. Within the sample, the nucleic acids for determination may be present in both single-stranded and double-stranded forms. They are DNAs and RNAs, more

particularly mRNAs, microRNAs, rRNA, and non-coding RNAs, and also the cDNAs generated therefrom by reverse transcription and employable for gene expression analysis and for analysis of the non-coding RNAs. Furthermore, the term "nucleic acid" also encompasses aptamer DNA and aptamer RNA and/or fragments of genomic DNA, which in particular may also have been methylated. The invention is further aimed at the quantification of DNA fragments obtained by immunoprecipitation and of fragments quantified by digital karyotyping and methylation-specific digital karyotyping. The invention likewise enhances the quantification of DNA fragments for metagenomic analyses and also for the analysis of fragments from subtractive DNA libraries. Likewise falling within the term "nucleic acids" are single-stranded or double-stranded polynucleotides of artificial (synthetic) origin, including those whose phosphate groups have been replaced by other atoms and/or whose bases have been modified, and which can be amplified by means of polymerization and which are sequenceable.

[0015] "Primer binding site or polymerase binding site (PB)" refers to a nucleic acid of known sequence which can serve preferentially as a binding site for a primer, for example for PCR, the amplification being effected using the corresponding primers and polymerases by enzymatic polymerization. In addition, it is also possible to utilize known sequences for polymerase binding to initiate an enzymatic amplification, as already present, for example, in known plasmids or other vectors. In such a case, using an insertion sequence, the RAT-TAG combinations can be incorporated into a vector, which gives rise to a nucleic acid construct in the context of the present invention that can be replicated by cloning. The polymerase binding site may, for example, be a promoter

GUARDANT HEALTH EX.1009.328

sequence or an RNA polymerase, for instance SP6 or T7-RNA polymerase, in which case linear amplification of the nucleic acid construct proceeding from the binding site can be effected by means of the corresponding polymerase.

[0016] By a "characterizing polynucleotide domain" is meant what is called a "TAG", namely a nucleic acid domain having a sequence which is characteristic of a nucleic acid which occurs in the sample and is to be determined quantitatively. Which part of a nucleic acid is selected as characterizing polynucleotide domain can in principle be selected freely, provided that the individual TAGs can be assigned to a particular nucleic acid present in the sample. If this assignment is ambiguous, it is possible to employ further methods in order to assure that there is no ambiguity, for example with real-time PCR with specific primers. As TAG, it is possible to utilize conserved portions of a gene sequence, but preference is given to utilizing characterizing polynucleotide domains having a sufficient sequence length. The characterizing polynucleotide domains should preferably have a sequence length of at least 8 nucleotides, preferably of at least 12 nucleotides, more preferably of at least 15 nucleotides. A characterizing polynucleotide domain may also be the entire nucleic acid to be determined. However, the domain should preferably not exceed a length of 50 nucleotides, preferably of 35 nucleotides, especially of 27 nucleotides. At the same time, it is also possible for comparatively long characterizing polynucleotide domains to be incorporated into a nucleic acid construct, although it is frequently sufficient to determine just a portion of this sequence after an amplification of the constructs.

GUARDANT HEALTH EX.1009.329

[0017] A "polynucleotide domain with random sequence" in the context of the present invention is a synthetically produced (artificial) "random tag" (RAT), the sequence of which permits unambiguous marking of the characterizing polynucleotide domain. The RATs are typically produced commercially by a "statistical" linkage of the individual nucleotides. This affords a mixture of RATs, with falling probability of the presence of RATs having the same sequence with rising sequence length and with decreasing number of RAT molecules in the mixture. The polynucleotide domains thus produced have a random sequence in the sense of the invention. A more complex approach is the production of RATs having a known sequence, in which case it is thus possible to produce RATs having a sequence which is guaranteed to be unique. RATs of this kind also constitute a "random sequence" in the sense of the present invention. It is also possible for RATs to be produced from a mixture of known, different sequences.

[0018] Single-stranded RATs may be produced or acquired as oligonucleotides. For production of double-stranded RATs, it is preferable to use single-stranded oligonucleotides having a random sequence (RAT) and a known sequence fraction, the known sequence fraction being at the 3′ end of the single-stranded oligonucleotide. The known sequence fraction is used to hybridize oligonucleotides having a complementary sequence as primers, which are converted with the aid of a polymerase (for example with what is called the Klenow fragment of DNA polymerase I) to a double-stranded oligonucleotide that contains a double-stranded RAT. Pure double-stranded RATs without further domains can be produced integration of a tagging enzyme site and a primer binding site, with elimination of the RAT with the aid of the tagging enzyme after second

strand synthesis. In addition, the single-stranded RAT can preferably be provided at one end with a known sequence which may contain particular polynucleotide domains such as a tagging enzyme site, restriction site, ligation site, etc. At the other end of the RAT, a primer binding site is then preferably inserted, which can also be employed for synthesis of the double strand with the Klenow fragment.

[0019] In order to provide RATs with suitable ligation sites (LS) complementary to overhanging restriction sites, it is possible to produce oligonucleotides containing a primer binding site followed by a RAT and the corresponding restriction site followed by a further RAT and a further primer binding site. A construct is obtained with the following structure: PB-RAT-restriction site-RAT-PB. After the production of the double-stranded construct, the oligonucleotides are digested with the appropriate restriction enzyme; two RATs are obtained in each case from one oligonucleotide and have been modified with protruding ends for the ligation.

[0020] In addition, the RATs can be produced by hybridization of two single-stranded oligonucleotides consisting of a random combination, which are hybridized under stringent conditions. In the case of RATs formed from known sequences, this is the preferred method.

[0021] By "nucleic acid constructs" are meant polynucleotides containing at least one characterizing polynucleotide domain which is characteristic of a nucleic acid which is present in the sample and is to be determined quantitatively (TAG), a polynucleotide domain having a random sequence (RAT) and at least one primer binding site or polymerase binding site (PB). In

GUARDANT HEALTH EX.1009.331

addition, further polynucleotide domains may be present in the nucleic acid constructs, for example linker sequences, ligation sites (LS), restriction enzyme sites or tagging enzyme binding sites (TES). In a preferred sequence of polynucleotide domains, the characterizing polynucleotide domains (TAG) and the polynucleotide domains having a random sequence (RAT) are joined covalently to one another. They may be joined to one another by direct ligation of the individual domains. Alternatively, the individual domains may also be connected to one another via linker sequences, in which case the linker sequence results from the ligation of corresponding overhanging ends. The TAG and RAT domains joined to one another are finally supplemented with at least one primer binding site or polymerase binding site (PB), which is connected, preferably in a covalent manner, either to the characterizing polynucleotide domain (TAG) or to the polynucleotide domain having random sequence (RAT). In a preferred embodiment, the TAG domains and the RAT domains are flanked by two different primer binding sites (PB, PB'). As well as direct linkage of the primer binding sites or polymerase binding sites to the respective ends of the TAG-RAT domain combination, another option is a linkage via a polynucleotide linker. It is also possible for the primer binding sites or polymerase binding sites (PBs) to be incorporated into the nucleic acid constructs by ligation. Alternatively, it is possible to use TAGs, but especially also RATs, which have already been provided with a primer binding site (PB) beforehand.

[0022] In an alternative embodiment, the nucleic acid fragments provided for enzymatic amplification may, as well as at least one characterizing polynucleotide domain (TAG), also contain two or more polynucleotide domains having random sequence (RATs). In this case,

one, two or more combinations of a TAG and a RAT may be present in a nucleic acid construct. The orientation of the TAG-RAT combinations in the construct is unimportant. In addition, individual TAG-RAT combinations may independently be flanked by a primer binding site or polymerase binding site (PB). However, it is also possible that two primer binding sites (PBs) include all the TAG-RAT combinations in a nucleic acid construct, or that a primer binding site or polymerase binding site (PB) is at one end of the TAG-RAT combinations.

[0023] Thus, particularly nucleic acid constructs having the following polynucleotide domain sequence are of particular interest:

a) PB-TAG-RAT,

b) PB-RAT-TAG,

c) PB-TAG-RAT-PB',

d) PB-RAT-TAG-RAT,

e) PB-RAT-TAG-RAT-PB',

f) PB-RAT-TAG-TAG-RAT,

g) PB-RAT-$(TAG)_n$-PB; where n = 2 to 50, preferably where n = 2 to 10,

h) PB-RAT-$(TAG)_n$-RAT-PB; where n = 2 to 50, preferably where n = 2 to 10,

i) PB-RAT-$(TAG)_n$; where n = 2 to 50, preferably where n = 2 to 10,

j) PB-RAT-TAG-RAT-TAG,

k) PB-TAG-RAT-TAG-RAT,

l) PB-RAT-TAG-RAT-TAG-PB',

m) PB-$(RAT-TAG)_n$-PB'; where n = 3 to 50, preferably where n = 3 to 10,

n) PB-RAT-TAG-PB'-PB''-RAT-TAG-PB''',

o) PB-RAT-TAG-PB'-PB''-RAT-TAG-PB'''-PB''''-RAT-TAG-PB'''',

p) PB-RAT-TAG-PB'-$(PB-RAT-TAG-PB')_n$-PB-TAG-RAT-PB'; where n = 0 to 10,

(PB, PB', PB'', PB''', PB'''', PB''''' = primer binding sites, where the primer binding sites preferably have different sequences, but may also have the same sequence).

[0024] Particular preference is given in the context of the present invention to nucleic acid constructs according to embodiments a), b), c), e) and g) and h).

[0025] Nucleic acid constructs may be in single- or double-stranded form, and a primer binding site in the case of single-stranded nucleic acid constructs is also understood to mean a sequence corresponding to a primer binding site on the opposite strand, whereas both the primer binding site and its corresponding sequence on the opposite strand are present in a double-stranded nucleic acid construct.

[0026] The nucleic acid constructs serve as templates for the enzymatic amplification thereof by a polymerase.

[0027] The nucleic acid constructs may also contain further oligonucleotide domains, for example linkers, ligation sites, restriction sites and tagging enzyme binding sites (TES).

[0028] In a further preferred embodiment, the nucleic acid construct or precursors of this construct contain one or more binding sites for a tagging enzyme (TE). A tagging enzyme site (TES) constitutes a binding site for a type-II or type-III restriction enzyme which cuts at a point far removed from its recognition site, preferably for EcoP15I, MmeI or BsmFI. Through use of a tagging enzyme, it is especially possible to produce nucleic acid fragments having a defined length which

GUARDANT HEALTH EX.1009.334

serves as characterizing nucleotide domain (TAG). The tagging enzyme site (TES) should be incorporated in such a way that the TAG required is present in the nucleic acid construct or any construct precursor even after the enzymatic cleavage. Preferably, the binding site for a tagging enzyme (TES) is at the distal end with respect to the PB sequence of the RAT; the binding of the TES into a nucleic acid construct or into a corresponding construct precursor is thus preferably effected in the domain sequence "PB-RAT-TES-nucleic acid". The product of the later cleavage of the nucleic acid construct with the TE is therefore a polynucleotide having the domain sequence PB-RAT-TES-TAG. Alternatively, it is also possible first to produce the TAG with the aid of the tagging enzyme and then the RAT is attached at either end of the TES-TAG domain sequence. If, for example, EcoP15I is used as tagging enzyme, the presence of two recognition sites oriented head-to-head (tagging enzyme sites) with the sequence CAGCAG is needed, which include the restriction site of the tagging enzyme and are preferably separated by fewer than 3000 Bp. The preferred domain sequence is therefore, in the case of use of a type III restriction enzyme, such as EcoP15I, "PB-RAT-TES-nucleic acid-TES". The products of the later cleavage of the nucleic acid construct with EcoP15I as TE are therefore a polynucleotide having the domain sequence PB-RAT-TES-TAG and a polynucleotide having the domain sequence TES-Tag, of which the PB-RAT-TES-TAG construct is preferably used for characterization of the nucleic acid. A major advantage in the case of use of EcoP15I is that particularly long TAGs having 25-27 base pairs are obtained.

[0029] In another preferred mode of application, both ends of the nucleic acid molecule to be characterized are provided with identical or different PB-RAT-TES

GUARDANT HEALTH EX.1009.335

constructs. The cleavage of the construct with the TE then results in two molecules having the domain sequence PB-RAT-TES-TAG, where the PB and TES domains may have an identical or different sequence. In this case, it is possible to use two PB-RAT-TES-TAG constructs for characterization of the nucleic acid molecule.

[0030] After cutting with the appropriate tagging enzyme, it is then possible to attach further polynucleotide domains, for example RATs and PBs, to the TAG obtained, so as to result in the following particularly preferred domain sequences in the resulting nucleic acid constructs:

a') PB-TES-TAG-RAT,

a'') PB-TAG-TES-RAT,

b') PB-RAT-TAG-TES,

b'') PB-RAT-TES-TAG,

c') PB-TAG-TES-RAT-PB',

c'') PB-TES-TAG-RAT-PB',

d') PB-RAT-TAG-TES-RAT,

d'') PB-RAT-TES-TAG-RAT,

e') PB-RAT-TAG-TES-RAT-PB',

e'') PB-RAT-TES-TAG-RAT-PB',

f) PB-RAT-$(TES)_m$-TAG-TAG-$(TES)_m$-RAT; where m = 0 or 1 independently of one another for each m, and at least one m = 1,

g') PB-RAT-TES-TAG-RAT-TAG,

g'') PB-RAT-TAG-TES-RAT-TAG,

k') PB-TES-TAG-RAT-TAG-RAT,

k'') PB-TAG-TES-RAT-TAG-RAT,

k''') PB-TAG-RAT-TES-TAG-RAT,

k'''') PB-TAG-RAT-TAG-TES-RAT,

l') PB-RAT-TES-TAG-RAT-TAG-PB',

l'') PB-RAT-TAG-TES-RAT-TAG-PB',

l''') PB-RAT-TAG-RAT-TES-TAG-PB',

l'''') PB-RAT-TAG-RAT-TAG-TES-PB',

m') PB-(RAT-(TES)$_m$-TAG)$_n$-PB'; where n = 3 to 50, preferably where n = 3 to 10 and m = 0 or 1 independently of one another for each m,

n') PB-RAT-(TES)$_m$-TAG-(TES)$_m$-PB'-PB''-RAT-(TES)$_m$-TAG-(TES)$_m$-PB''; where m = 0 or 1 for each m independently of one another, and at least one m = 1,

o') PB-RAT-(TES)$_m$-TAG-(TES)$_m$-PB'-PB''-RAT-(TES)$_m$-TAG-(TES)$_m$-PB'''-PB''''-RAT-(TES)$_m$-TAG-(TES)$_m$-PB'''''; where m = 0 or 1 for each m independently of one another, and at least one m = 1,

p') PB-RAT-(TES)$_m$-TAG-(TES)$_m$-PB'-(PB-RAT-(TES)$_m$-TAG-(TES)$_m$-PB-(TES)$_m$-TAG-(TES)$_m$-RAT-PB' where n = 0 to 10 and where m = 0 or 1 for each m independently of one another, and at least one m = 1,

(PB, PB', PB'', PB''', PB'''', PB''''' = primer binding sites, where the primer binding sites preferably have different sequences, but may also have the same sequence).

[0031] The nucleic acid constructs can be produced by means of circular nucleic acid construct precursors, preferably by modifying at least one end of a nucleic acid with a marker and optionally modifying the other end likewise with a marker or adapter, followed by circularization. Preferably, at least one of the markers or adapters present also has a tagging enzyme binding site (TES); more preferably, two TES are present, especially for the tagging enzyme EcoP15I which cuts off a TAG on either side of the circular construct, forming a linear nucleic acid construct in the sense of the present invention that contains two TAGs at either end.

[0032] Further preferred nucleic acid constructs which can be produced, for example, from circular construct

precursors and which are of particular interest for the simultaneous quantification of both ends of nucleic acid molecules because they allow recognition of the origin of TAGs of the same molecule are:

q) TAG-TES-RAT-PB-RAT-TES,
r) TAG-TES-RAT-PB-PB'-RAT-TES,
s) PB''-TAG-TES-RAT-PB-RAT-TAG,
t) PB''-TAG-TES-RAT-PB-RAT-TAG-PB', or
u) PB''-TAG-TES-RAT-PB-PB'-RAT-TAG-PB'',
v) TAG-RAT-PB-RAT,
w) TAG-RAT-PB-PB'-RAT,
x) PB''-TAG-RAT-PB-RAT-TAG,
y) PB''-TAG-RAT-PB-RAT-TAG-PB',
z) PB''-TAG-RAT-PB-PB'-RAT-TAG-PB''',
aa) PB-TAG-RAT-TAG,
bb) PB-TAG-RAT-TAG-PB',

(PB, PB', PB'', PB''' = primer binding sites, where the primer binding sites preferably have different sequences, but may also have the same sequence).

[0033] All the nucleic acid constructs presented in the present description of the invention may, as well as the domain sequences of the invention, have further sequence sections, for example linkers, restriction sites, ligation sites, etc., although these are generally not specified explicitly in the domain sequence of the constructs.

[0034] The polynucleotide domains having random sequence (RAT) are suitable for marking characterizing polynucleotide domains (TAGs). By a "marker" is consequently meant polynucleotides containing a primer binding site and a RAT but no TAG. In addition, markers may contain further oligonucleotide domains, for example tagging enzyme binding sites (TES) and linker and ligation sites (LS). A ligation site is the end of

GUARDANT HEALTH EX.1009.338

the marker intended for ligation, which is in a distal position relative to the PB.

[0035] For the production of the nucleic acid constructs intended for amplification, it is additionally advisable also to include particular oligonucleotide domains as well as the markers in an "adapter". Adapters in the sense of the present invention are always provided with a primer binding site and may contain further oligonucleotide domains, especially tagging enzyme binding sites, linkers or ligation sites, but no RATs or TAGs.

[0036] The markers and adapters preferably contain different primer binding sites which are to be used preferentially in the highly parallel sequencing methods according to the manufacturers' instructions. These are at the distal ends of the markers or adapters from the TAG.

[0037] The individual oligonucleotide domains, for example PB, TAG and RAT, and the markers or adapters, may additionally be provided by the standard prior art methods with further known modifications which, for example,

- enable binding to a solid phase, for instance by biotinylation of the oligonucleotides and binding to a solid phase provided with streptavidin, or contain "scavenger sequences". "Scavenger nucleotide sequences" utilized may be sequences bound, for example, to a solid phase, which can hybridize with their corresponding opposite strands present in solution,
- prevent ligation on one side of an oligonucleotide, for example amino-C7 modifications or "reverse nucleotides" (nucleotides with 3'-5 orientation rather than 5'-3' orientation), in

GUARDANT HEALTH EX.1009.339

which case the end to be protected from ligation is the distal end of the marker or adapter from the TAG,

- enable or facilitate ligation on one side of an oligonucleotide, for example by phosphorylation of the 5' end of the corresponding oligonucleotide, or, for example, through the generation of overhanging restriction sites ("sticky ends"),
- facilitate the hybridization or frustrate the dehybridization of the double strand, for example through the use of "locked nucleotide acids",
- include restriction sites.

[0038] These modifications may be integrated into the single-stranded oligonucleotides. Adapters may be hybridized with complementary oligonucleotides to give double-stranded oligonucleotides. Markers may optionally likewise be produced, but the marker is preferably produced in double-stranded form through the use of a polymerase, especially by means of the Klenow fragment of DNA polymerase I as described above.

[0039] With the aid of markers, it is possible to unambiguously characterize nucleic acid fragments having a sequence characteristic of a nucleic acid which occurs in the sample (TAG), meaning that every RAT-TAG combination is highly likely to be unique. In order to undertake such an unambiguous characterization, the nucleic acid domain having random sequence (RAT) should preferably be long enough to be able to provide an excess of markers having different RATs in relation to the number of nucleic acids that occur in the sample. Preference is given to using markers containing RATs and having more than twice the amount relative to the nucleic acid fragments or nucleic acids to be marked. In the individual case, however, a number of markers corresponding to the

number of nucleic acids to be determined may also be sufficient, especially when all the RATs are guaranteed to be of different sequence. Preferred sequence lengths for the RATs present in the nucleic acid fragments are between 2 and 100 nucleotides, better between 4 and 50 nucleotides; particularly preferred RATs have a total length between 4 and 15 nucleotides. The entire random sequence may be present in one marker domain or be divided between several marker domains or between domains of different markers in a nucleic acid construct. With the described total lengths of the nucleic acid domains having random sequence (RATs), it is theoretically possible to produce a large number of markers having different RATs, in which case the marker ensemble used typically contains only a small portion of the conceivable RATs that arise from the sequence length, such that the probability that RATs that coincidentally have identical sequences will occur in two markers is very low. Consequently, the sequence length of the RATs can be matched individually to the number of characterizing nucleic acid fragments to be marked. For example, for a gene expression analysis in which the mRNAs present in a tissue sample are to be determined quantitatively, preference is given to using marker mixtures having more than 10^2 markers, more preferably between 10^4 and 10^8 markers.

[0040] Even if individual markers should indeed contain identical RATs, this only distorts the quantitative determination if they should become bonded to a TAG of identical or similar sequence. Since all the individual TAG-RAT combinations for the individual nucleic acids that occur in the sample and are to be determined quantitatively should be present only once in a mixture prepared for amplification, it is consequently possible to identify sequence-identical combinations of a characterizing polynucleotide domain

(TAG) and a polynucleotide domain having random sequence (RAT) as copies of a characterizing polynucleotide domain of a nucleic acid originally present in the sample.

[0041] Since polymerization errors can also occur during the amplification of the nucleic acid constructs and the determination of the nucleic acid sequence is not free of errors either, it is advantageous also to eliminate sequence-similar RAT-TAG combinations in the evaluation. What can be considered as "sequence-similar" combinations can in principle be fixed freely.

[0042] The frequency with which such errors occur depends on the length of the TAGs and RATs, the amplification conditions and the sequencing methods. "Sequence-similar" copies are preferably considered to be RAT-TAG sequences having a maximum of one (1) deviation for every 10 nucleotides; preferably, a sequence-similar copy is defined as RAT-TAG combinations having not more than one deviation for every 15 or 20 nucleotides. For example, sequence-similar RAT-TAG combinations having a sequence length of 40 base pairs should have a maximum of 4 sequence deviations, unless working under conditions where errors are tolerated. If amplification errors and sequencing errors can be substantially eliminated, it is advantageous to lower the error limit for "sequence-similar" RAT-TAG combinations.

[0043] **Fig. 1** shows an example of four RAT-TAG combinations having an identical TAG. Combinations 1, 2 and 3 are different; combination 4 is a copy of 3. The TAG thus occurs three times in the sample. For quantification, only one combination is counted, for example 3, while combination 4 is eliminated from the data set.

GUARDANT HEALTH EX.1009.342

[0044] The nucleic acid containing the TAG is preferably in double-stranded form, for example as cDNA or DNA, and subsequently ligated to the double-stranded construct(s) (markers, adapters). For linear amplification of RNA, the nucleic acid to be amplified may, however, also be single-stranded. Single-stranded RNA can be provided with markers with the aid of RNA ligase, for example the T4 RNA ligase, prior to amplification.

[0045] There follows a description of some strategies for production of the nucleic acid constructs from double-stranded DNA. Because of the modular character of the individual method steps for production of the constructs, it is also possible in an analogous manner to produce nucleic acid constructs with different domain sequences, or to obtain identical nucleic acid constructs with an alternative sequence of method steps.

[0046] The individual method steps which are advantageous for production of the nucleic acid constructs are

- the binding of single- or double-stranded oligonucleotides, especially of markers and adapters, or of the nucleic acids to a solid phase, and the elimination thereof,
- the ligation of the individual markers, adapters and the nucleic acids and
- the digestion of nucleic acid strands with a restriction and/or tagging enzyme.

[0047] In a preferred embodiment for production of double-stranded nucleic acid constructs which comprise, for example, TAGs from cDNA or genomic DNA and are to be determined quantitatively, the double-stranded

nucleic acids are provided with a marker or adapter which can be bound to a solid phase.

[0048] Binding to a solid phase has the advantage that, for example, nucleic acids that have not been ligated or are not required can be removed after a restriction digest, or nucleic acids required can be removed from the solid phase. However, the binding of the nucleic acids to a solid phase is also dispensable for some methods.

[0049] Preferably, markers or adapters are biotinylated for this purpose and can be bound to a streptavidin-coupled solid phase. The nucleic acids to be examined in a mixture may be ligated to those markers or adapters which have been biotinylated at the opposite end from the end intended for ligation (ligation site = LS). This can be accomplished, for example, after digestion of the DNA to be quantified, with a restriction enzyme at a restriction site or else at blunt ends of the nucleic acid. Biotinylated markers or adapters, in the case of cDNA, can preferably also be accomplished through the use of biotinylated single-stranded oligonucleotides which have a poly-T sequence at the 3′ end and are integrated by first- and second-strand synthesis, beginning at the 3′ end of the cDNA (the 3′ end of the cDNA is considered to be the end corresponding to the 3′ end of the mRNA. It is characterized by an oligo-A/T domain).

[0050] In a preferred embodiment, the nucleic acid (cDNA, DNA) is digested with one or more frequent-cutting restriction enzymes. Frequent-cutting restriction enzymes are, for example, NlaIII, Hsp92II, FatI, BfaI, MaeI, XspI, HpyCH4IV, MaeII, TaiI, TscI, AluI, TaqI, BfuCI, Bsp143I, BstENII, DpnII, Kzo9I, MboI, NdeII, Sau3AI, BstKTI or Csp6I. These enzymes

produce nucleic acid fragments having an average length of 200 to 300 base pairs. The nucleic acids in the mixture that are to be determined quantitatively can be bound to a solid phase before or after this digest, for example after ligation with biotinylated markers or adapters. As an alternative to the digest, the nucleic acids in the mixture that are to be determined can be comminuted by shear prior to binding to the solid phase.

[0051] Fragments not bound to the solid phase can be removed by washing.

[0052] The bound fragments can be provided again, preferably by ligation, with a marker or adapter which should not be biotinylated. If the nucleic acids to be determined quantitatively have been digested with a restriction enzyme beforehand, markers or adapters may be provided with the ends corresponding to the restriction site and ligated with the digested nucleic acids to be determined quantitatively. The other end of the adapter or marker can be protected from ligation, for example by the incorporation of an amino-C7 modification at the distal end of the marker or adapter from the LS. After mechanical shear, the nucleic acids to be determined quantitatively, after filling in or removing protruding individual strand ends ("sticky ends"), for example with the Klenow fragment, can likewise be provided with appropriate non-biotinylated markers or adapters.

[0053] Thereafter, the nucleic acids in the mixture that are to be determined quantitatively are preferably in the form of:

PB-TAG-RAT-PB',

PB-RAT-TAG-RAT or

PB-RAT-TAG-RAT-PB'

and can be amplified, for example, by PCR and sequenced, for example, by highly parallel sequencing methods.

[0054] For quantitative analysis of gene expression, it is generally sufficient to use the cDNA ends (Torres T. T. et al., Gene expression profiling by massively parallel sequencing, Genome Res. 2008 18:172-177) as TAGs. For this purpose, mRNAs are converted to a cDNA, for example using a biotinylated poly-T primer (PB1 = adapter), and bound to a solid phase. This is followed by digestion with a frequent-cutting restriction enzyme (anchoring enzyme). This is followed by ligation with a PB'-RAT marker containing a second primer binding site (PB2) in which the distal end from the ligation site is protected from ligation. The resultant nucleic acid construct is consequently PB2-RAT-TAG-PB1. The resultant nucleic acid constructs are amplified by means of PCR and subsequently sequenced.

[0055] A further way of obtaining nucleic acid constructs particularly suitable for highly parallel sequencing methods is the use of biotinylated markers or adapters, the binding thereof to a solid streptavidin phase and the obtaining of single-stranded constructs after denaturing. The markers or adapters are 5'-phosphorylated at a distal position from the primer binding site and protected from ligation at the primer binding site end, for example with an amino-C7 modification.

[0056] The constructs c, d and e that are of particular suitability for pyrosequencing are obtained by ligating the TAG, which may, for example, be a cDNA or a genomic DNA fragment, with markers and adapters, for example by means of specific restriction sites or blunt ends, with one of the individual strands of the

marker formed from two individual strands, for example PB-RAT-LS or adapter, for example PB'-LS, in biotinylated form. Ligation additionally gives rise to the unwanted by-products PB'-TAG-PB' and PB-RAT-TAG-RAT-PB. These have either been biotinylated on both strands or not biotinylated at all.

[0057] After binding to a streptavidin phase, the unbound fragments are first removed by washing. If the double-stranded constructs are then denatured, the fragments having two biotinylated adapters remain on the solid phase. Only those nucleic acid constructs having the desired formation go into solution as a single strand and can thus be recovered. The single-stranded constructs can be produced in double-stranded form again with the aid of a polymerase. This method is versatile and can in principle be used for all double-stranded DNA nucleotides.

Obtaining TAGs by using a tagging enzyme:

1) Obtaining a tag of a cDNA provided with a RAT:

[0058] In a further embodiment, the nucleic acids in a mixture that are to be determined quantitatively, as described above, are provided with nucleic acid constructs (markers and adapters) which contain a tagging enzyme binding site (TES) and are bound to a biotin molecule at one end (e.g. TES-RAT-PB-biotin). It is thus possible to bind the nucleic acids to a solid phase, for example streptavidin-coupled magnetic particles or "beads". After digestion with one or more frequent-cutting restriction enzymes or shearing, the construct, once it has been bound to a solid phase, can be cut with a tagging enzyme, for example BsmFI or MmeI.

GUARDANT HEALTH EX.1009.347

[0059] It should be mentioned at this point that fragments which are eliminated by a tagging enzyme will have an equal length if the markers and adapters used were likewise of the same length and the same sequence of domains is present in all molecules, as is preferably the case according to the present invention. The nucleic acid fragments eliminated by the tagging enzyme can thus be separated and isolated in a simple manner on the basis of size, for example by electrophoretic methods or by HPLC. Even after ligation with a marker or adapter of a particular size, the ligation products can be isolated on the basis of size. It is thus also possible to isolate the corresponding specific amplification products.

[0060] If a type III enzyme, for instance EcoP15I, is used as tagging enzyme, it is necessary beforehand to ligate an adapter or marker containing a second TES to the free end of the nucleic acid strands fixed to the solid phase. After digestion with the tagging enzyme and the removal of the eliminated products not bound to the solid phase, it is possible to fill in any protruding ends, for example with the Klenow fragment, or to eliminate them with a single-strand-specific exonuclease, in order that the tagging enzyme restriction site has blunt ends. It is then possible to bind markers or adapters to the fixed nucleic acid strand. Alternatively, it is possible to use adapters or markers which have been provided with "sticky ends" and are complementary to the end that results from the tagging enzyme.

[0061] The products which arise are preferably, according to the positioning of the marker or adapter,

PB-TAG-TES-RAT-PB'

or PB-RAT-TAG-TES-RAT-PB'.

GUARDANT HEALTH EX.1009.348

[0062] Nucleic acid constructs can be produced either using a solid phase or in solution. In a preferred embodiment, for this purpose, the nucleic acids or nucleic acid fragments present in a sample are ligated at both ends with markers or adapters. The ends may be defined beforehand by a restriction enzyme and provided with appropriate markers or adapters. The resultant construct has the following domain sequence:

PB-nucleic acid-PB

or

PB-RAT-nucleic acid-RAT-PB.

[0063] If this construct is digested with a frequent-cutting enzyme, the result in many cases is further restriction sites in the nucleic acid. These can be used for the ligation of a further adapter or marker. This results in the following products:

PB-RAT-TAG-PB

or

PB-TAG-RAT-PB.

[0064] If, in a first step, adapters or markers containing tagging enzyme recognition sites are used, the result is:

PB-RAT-TES-nucleic acid-TES-RAT-PB'

or

PB-TES-nucleic acid-TES-PB'.

[0065] Thereafter, after digestion with the tagging enzyme, two nucleic acid fragments containing a TAG can be eliminated from the nucleic acid in each of the nucleic acid strands obtained. Preferably, a marker or adapter is attached to the restriction sites of the tagging enzymes. Among others, the following nucleic acid constructs will arise, which can be isolated on the basis of their size, for example by means of gel electrophoresis or HPLC:

PB-RAT-TES-TAG-PB'
and
PB'- RAT-TES-TAG-PB
or
PB-TES-TAG-RAT-PB'
and
PB'- TES-TAG-RAT-PB.

[0066] As a result of the formation of two nucleic acid constructs for each nucleic acid present in the sample, an internal control is possible in the quantitative determination of the nucleic acids.

[0067] It is apparent that the nucleic acid constructs which are required for amplification and contain the characteristic TAG-RAT combinations can be produced in various ways. The nucleic acid constructs can be formed in a modular manner; for example, it is possible to combine the individual method steps for binding or production of individual polynucleotides with the different functional domains (TAG, RAT, PB, TES) in a simple manner in order to obtain the desired nucleic acid constructs. As a result, the method of the invention is versatile and is not restricted to a particular way of processing the nucleic acids present in the sample.

[0068] There follows an elucidation in detail, by way of example, of the individual method steps which can be employed in a modular manner.

1. Preparation of an RNA-containing sample for production of cDNA-TAGs:

[0069] With the aid of a commercial kit, for example comprising Trizol reagent (Invitrogen Corp., Carlsbad, USA), complete RNA is obtained from a biological

sample. The mRNA is isolated from the complete RNA with a commercial kit, for example "Oligtex-Midi-Kit" (Qiagen N.V., Venlo, the Netherlands). The cDNA is produced by reverse transcription using a 5'-biotinylated poly-T oligonucleotide, for example with the "cDNA synthesis system" (Invitrogen Corp.). The product is converted to a double-stranded DNA ("cDNA synthesis system", Invitrogen Corp.) and dissolved in a suitable buffer. In a particularly preferred variant of the method, the poly-T oligonucleotide used should contain the recognition sequence for EcoP15I CAGCAG or CTGCTG,
for example 5'-biotin-PB-CAGCAGTTTTTTTTTTTT
TTTTT-3' (SEQ ID No. 1).

2. Enzymatic digestion of nucleic acids present in the sample:

[0070] Double-stranded DNA is digested in a reaction solution with a frequent-cutting restriction enzyme ("anchoring enzyme"), e.g. NlaIII, in buffered solution. After the digestion, the resultant DNA is extracted, precipitated with alcohol and taken up in a suitable buffer. Thereafter, the DNA solution is admixed with streptavidin-coated particles. The particles are incubated at room temperature in suitable buffer, in order to enable binding of the biotinylated cDNA with the streptavidin-coated particles. The DNA bound to the particles is washed and taken up in a suitable buffer.

3) Production and ligation of polynucleotide domains, adapters and markers:

[0071] Adapters are assembled from two complementary single-stranded oligonucleotides. The two single-stranded oligonucleotides are dissolved in a suitable

buffer and hybridized with one another to form a double strand. The marker containing the RAT is likewise at first synthesized in single-stranded form and a double strand of this template is synthesized as described above with the aid of the Klenow fragment. The adapter and marker are protected from unwanted ligation on the distal side from the LS through the incorporation of, for example, an amino-C7 modification of the 3′ side of the corresponding oligo, while ligation is enabled at the ligation site through 5′-phosphorylation. The double-stranded marker or adapter can then be joined with a ligase, e.g. T4 ligase (Invitrogen Corp.), in a suitable buffer, covalently to a 3′ end of a different polynucleotide or a nucleic acid from the sample or a fragment thereof that contains the TAG and may be bound to a solid phase.

[0072] If the marker and adapter are to be ligated, they are preferably added in a sufficiently large amount, such that all the free ends of the nucleic acid strands that are to be formed are also ligated with the polynucleotides.

4) Cutting with the tagging enzyme:

[0073] In a suitable buffer, the nucleotides which are to be quantified and have been provided with an adapter or marker containing a TES are incubated with a tagging enzyme, for example BsmFI, MmeI, EcoP15I.

[0074] The eliminated nucleic acid fragments are isolated either by electrophoresis or HPLC, or are (in the case of use of corresponding markers or adapters) bound to a solid phase. If further markers or adapters are to be bound to the tagging enzyme restriction site and the restriction site is not a blunt restriction site, the overhanging end can be filled in or

eliminated with an exonuclease. For this purpose, for example, it is possible to use the Klenow fragment of the DNA polymerase.

5) Amplification:

[0075] For replication of the nucleic acid constructs, preference is given to using PCR-based amplification methods. Alternatively, it is also possible to use unidirectional amplification or cloning methods for replication of the nucleic acid constructs.

6) Sequencing:

[0076] The sequence of individual nucleic acid fragments obtained by amplification in a mixture is preferably determined by suitable sequencing methods. Such methods are applied commercially, for example "454" picoliter methods, with the GSFLX system (Roche Diagnostics, Germany), Solexa methods (Illumina, Inc., San Diego, USA); SOLid methods (Applied Biosystems, Inc., Foster City; USA); HeliScope™ Single Molecule Sequencer (Helicos Corp., Cambridge, USA). With these methods, it is nowadays possible to sequence up to several million sequences in parallel. Parallel sequencing of a mixture of amplified nucleic acid constructs is possible without further separation and without an intermediate cloning step. However, it is also possible to assemble the nucleic acid fragments obtained to form concatemers which can subsequently be cloned, processed and sequenced by conventional methods (for example according to Sanger).

[0077] Since the quantitative evaluation of the nucleic acid constructs produced is effected by sequencing, the portion of the constructs to be sequenced, in a preferred embodiment, should not exceed

GUARDANT HEALTH EX.1009.353

a length of 400 nucleotides. Preferred nucleic acid constructs therefore have a length between 15 and 150 base pairs, more preferably between 25 and 75 base pairs. However, it is also possible for longer sequences to be sequenced, up to the complete nucleic acid sequences.

Brief description of the figures which show individual possible embodiments of the invention by way of example:

[0078] **Fig. 1** shows an example of four RAT-TAG combinations with an identical TAG.

[0079] **Fig. 2** shows, in schematic form, the production of nucleic acid constructs from mRNA for quantitative analysis of the gene expression.

[0080] **Fig. 2**, steps 1.1 to 1.3, shows linear amplification using a T7 promoter (PB'). For this purpose, a marker having the domain sequence PB'-PB-RAT-poly-T where PB' contains the promoter sequence for a T7 or SP6-RNA polymerase is hybridized with the poly-A end of the mRNAs in the sample. The free 3' end of the poly-T sequence then serves as a starting point for a reverse transcription (first-strand synthesis: **fig. 2**, step 1.1). By second-strand synthesis, for example using oligohexamers, a double-stranded cDNA is produced therefrom (step 1.2). Using the T7 polymerase, linear amplification can then be effected from the cDNA for production of antisense RNA (aRNA) (step 1.3). The resultant aRNA can then be transcribed using the "random hexamer" as primer by reverse transcription to give the corresponding first DNA strand (step 1.4); the corresponding amplified single-stranded cDNA is obtained. With the aid of the primer binding site (PB), it is then possible to amplify the corresponding first

GUARDANT HEALTH EX.1009.354

DNA strand (step 1.6.1), and it is also possible to use biotinylated primer binding sites (step 1.6.2). The resultant double-stranded constructs can then be analyzed further, for example as shown in **fig. 3** (step 3-5).

[0081] **Fig. 2**, steps 2-4, shows amplification of mRNA by what is called the SMART method.

[0082] **Fig. 3** shows, in schematic form, the production of nucleic acid constructs from genomic DNA using the example of digital karyotyping. For this purpose, the genomic DNA present in a sample is digested using a methylation-insensitive restriction enzyme (**fig. 3**, step 1); the fragments obtained preferably have overhanging ends, such that it is possible to ligate markers of the biotin-PB-RAT form onto the corresponding restriction sites in the genomic DNA, and these markers have appropriate ligation sites at distal positions from the primer binding site (step 2). The resultant nucleic acid construct precursors can then be digested with a further restriction enzyme (step 3). After binding of the marker-containing fragments to a streptavidin matrix (e.g. streptavidin-coated magnetic particles), it is then possible to ligate an adapter containing a second primer binding site (PB') onto the bound fragments via the corresponding overhanging ends ("sticky ends"). The nucleic acid constructs thus obtained can then be amplified and sequenced. In the case of methylation-specific DK, the first enzyme is replaced by a methylation-sensitive enzyme.

[0083] In an alternative embodiment, which is shown in **fig. 4**, steps 5-8, it is also possible to use markers which contain tagging enzyme binding sites and have been biotinylated in the present case and adapters for the formation of the nucleic acid constructs. In that

case, the nucleic acid constructs shown in step 5 are obtained. By adding the tagging enzyme, short TAGs are then cut out of the nucleic acid (step 6). If these have protruding ends, the resultant fragments are then filled in, for example, with the Klenow fragment. It is then possible to ligate the adapter-containing fragments present in solution with markers to the corresponding nucleic acid construct via the blunt ends thereof (step 7.1) and then to amplify them. The amplified nucleic acid construct can then be purified, for example by electrophoresis, and sequenced. It is also possible for the fragment bound to the solid phase to be processed further to give the desired nucleic acid construct by ligating the blunt end with an adapter containing a second primer binding site (step 7.2). The bound nucleic acid construct can be amplified directly and subsequently sequenced. Alternatively, it is possible to ligate the fragments which are present in solution and contain the adapter and a TAG and the bound fragments containing the marker and a TAG to one another to form ditags (step 6.2) prior to the amplification and sequencing of the resultant nucleic acid construct.

[0084] The applications of the invention described hereinafter demonstrate the various possible uses of the invention. The following are described in detail:

1) RAT marking of nucleic acids prior to the replication thereof by linear amplification:

2) Constructs for the quantification of cDNA,

2.1.) Constructs for the quantification of 3 ends of cDNA,

2.2.) Constructs for the quantification of 5′ ends of cDNA,

2.3.) Constructs for the simultaneous quantification of 5′ and 3′ ends of cDNA,

2.4.) Constructs for the simultaneous quantification of 3' and 5' ends of the same cDNA with the aid of a circularization method,

3) Constructs for the quantification of cDNA which have not been produced from poly-A-containing RNA,

4) Constructs for the quantification of fragments of genomic DNA,

4.1.) Constructs for the quantification of previously bisulfite-converted genomic DNA,

4.2.) Constructs for the quantification of both ends of fragments of genomic DNA with the aid of a circularization method.

1) RAT marking of nucleic acids prior to the replication thereof by linear amplification:

[0085] The aim of "linear amplification" is to replicate nucleic acids, for example in order to obtain sufficient material for a gene expression study, in such a way that the comparative composition of a nucleic acid sample is preserved. There exist various methods for this purpose. For instance, first of all, antisense RNA (aRNA) is produced in a linear manner, for example, from an mRNA to which a single-stranded marker containing a RAT has been bonded, with the aid of T7 polymerase or SP6-RNA polymerase (US 6,916,633) or with the aid of an RNA primer which a T7 promoter Dafforn et al., Biotechniques 2004 Nov.; 37(5): 854-7. However, sequence-related deviations in the copy number also occur in the case of "linear amplification" of RNA (e.g. Caretti et al. 2008, J. Cellular Biochemistry 103:556-563), which can be recognized by incorporation of a single-stranded RAT. In an alternative method, it is possible, for example, to bind a single-stranded marker containing a RAT to the 5' end of mRNAs present in the sample. This is then followed by the linear

GUARDANT HEALTH EX.1009.357

amplification, for example, proceeding from the 3' end with poly-T primers using a reverse transcriptase.

2) Constructs for the quantification of cDNA

[0086] Linear amplification is not a necessary prerequisite for the quantification of cDNAs which have been produced from mRNA having a 3'-poly-A end. However, it is an option when the mRNA is available only in small amounts. Therefore, what are described first of all are constructs which enable the linear amplification of the cDNA via an RNA step. There then follow descriptions of methods which proceed from nucleic acids available in sufficiently large amounts, whether they have been generated by amplification or not. Thereafter, there are descriptions by way of example of methods which enable the quantification of characterizing nucleic acid domains (TAGs) from nucleic acids using RATs.

2.1. Constructs for the quantification of 3' ends of cDNA

[0087] For amplification, the mRNA is first converted to cDNA with the aid of an oligo-dT-containing primer. For the later amplification with a polymerase, for example the SP6, T3 or T7 RNA polymerase, this primer also contains the promoter sequence for the polymerase. The later double-stranded cDNA then has the domain structure "cDNA-oligo-A/T-promoter". Changes in the nucleic acid composition can be recognized when a RAT is integrated between the promoter and the DNA to be amplified. The resulting domain structure in this case is

"cDNA-oligo-A/T-RAT-promoter",

where the T7 promoter sequence may also simultaneously serve as primer binding site. However, it is also

possible to incorporate a separate primer binding site (PB) as well as the promoter (polymerase binding site), such that the following domain structure is obtained: "cDNA-oligo-A/T-RAT-PB-promoter".

[0088] For quantification without prior direct amplification, for example with the aid of a polymerase, for example the SP6, T3 or T7 polymerase, a construct is chosen which, rather than the particular promoter, contains a primer binding site (PB) and/or a TES. A corresponding domain structure is then: cDNA-oligo-A/T-TES-RAT-PB.

[0089] The two preferred methods which follow can likewise be used to quantify TAGs which have been obtained from the 3' ends of a cDNA and contain the poly-A portion.

[0090] For the quantification of cDNA ends in a preferred method, mRNA is transcribed to cDNA with a poly-T-containing marker poly-T-RAT-PB-biotin by means of first and second strand synthesis. The cDNA is then digested with one or more frequent-cutting enzymes (anchoring enzymes). The marker-containing fragments are then bound to a streptavidin-modified matrix, for example magnetic particles. The unbound fragments are removed by washing. This is followed by ligation with an adapter containing a ligation site at the distal end from the primer binding site, which is protected from ligation by an amino-C7 modification, utilizing the overhanging ends for the ligation. After removal of the non-ligated adapters, the resultant nucleic acid constructs can be amplified and sequenced. Alternatively, it is also possible to use a marker rather than the adapter in the method described above. In this case, the mRNA need not be transcribed with a RAT-containing poly-T oligonucleotide.

GUARDANT HEALTH EX.1009.359

[0091] One of the preferred methods for the quantitative analysis of gene expression is referred to as the "super-TAG" method. For this purpose, mRNA is transcribed to cDNA using a 3'-biotinylated poly-T oligonucleotide with an EcoP15I binding site (EcoP15ITES) upstream of the poly-T sequence. This is followed by digestion with a frequent-cutting restriction enzyme (anchoring enzyme), for example with NlaIII. The cut cDNA is bound to streptavidin-coated magnetic particles by the biotin group. Alternatively, the oligo-dT may already be bound to the particles prior to the cDNA synthesis, or digestion with the anchoring enzyme may not take place until after binding to the particles. The unbound fragments are removed by washing.

[0092] In the next step, ligation is effected with a double-stranded PB'-EcoP15ITES oligonucleotide (adapter). The resultant nucleic acid strands are then incubated with EcoP15I and the fragments eliminated are isolated by a gel electrophoresis. The adapter-TAG constructs can then be ligated with a marker. The resultant nucleic acid constructs have the following domain sequence:
PB'-EcoP15ITES-TAG-RAT-PB.

[0093] This is followed by PCR amplification and the sequencing of the amplified constructs.

2.2. Constructs for the quantification of 5' ends of cDNA

[0094] For the exact quantification of the 5' ends of cDNAs, RATs may be attached to the 5' CAP sequence of the mRNA. The 5' CAP sequence consists of a guanine nucleotide bound to the mRNA by an unusual 5'-to-5'

triphosphate binding. For the attachment of a RAT, it is possible to proceed, for example, analogously to the production of full-length cDNA (Clepet et al. Improved full-length cDNA production based on RNA tagging by T4 DNA ligase, Nucleic Acids Research, 2004, vol. 32, no. 1 e6). In this case, RNAs are treated, for example, with alkaline phosphatase. Transcripts having an intact 5' CAP sequence are not dephosphorylated by these enzymes, whereas fragmented mRNA molecules and non-coding RNAs which do not have such a CAP sequence are dephosphorylated and end not with the phosphate group but with a 5'-OH group, to which the RNA polymerase cannot bind any RNA oligonucleotides. In the second step, the CAP sequence is then removed enzymatically, releasing the 5'-phosphate group which is then available for the ligation of an RNA oligonucleotide. This oligonucleotide of known sequence serves later as primer binding site (PB) for the second strand synthesis of the cDNA. In order to insert a RAT into this construct, the sequence of the RNA oligonucleotide should be chosen such that the later RNA construct has the domain sequence 5'-PB - RAT - mRNA-3'. It is optionally possible to insert a (T7 polymerase) promoter sequence into this construct, which can then serve as polymerase binding site and simultaneously as primer binding site (PB). The corresponding domain structure is then
5'-(T7)promoter - RAT - mRNA-3'.

[0095] Another method for providing the 5' end of an RNA with a primer is the use of the "Switch Mechanism At the 5'-end of Reverse Transcript" (SMART, TAKARA, Seta Otsu, Japan). In this case, the first strand synthesis of the cDNA results in an overhang, on the 3' side of the newly formed DNA strand, which is formed by the reverse transcriptase and consists of a plurality

GUARDANT HEALTH EX.1009.361

of (usually three) cytosine bases (see also **fig. 2**, steps 2 to 4).

[0096] This overhang is available as a binding site for an oligonucleotide primer. It is possible here, for example, in order to introduce the RAT, to use an oligonucleotide having a primer binding site at the 5′ end, followed by a RAT which concludes in turn with three successive guanine bases at the 3′ end, as primer for the second strand synthesis of the cDNA.

[0097] For many purposes, it is sufficient merely to obtain a TAG from the 5′ end of a cDNA which can then serve for quantification of the original mRNA. For this purpose, as described above, a nucleic acid construct comprising at least a PB (or a promoter sequence, for example of SP6, T3 or T7), a RAT and a TES is bound to the 5′-phosphate group released after the dephosphorylation and removal of the CAP site. The corresponding single-stranded RNA construct then preferably has the following form:
5′-biotin-PB/promoter-RAT-TES-mRNA.

[0098] At the same time, it is possible to bind a DNA construct containing a poly-T sequence to the poly-A sequence of the 3′ end of the mRNA by "hydrogen bonding" between the poly-A and the oligo-T sequence of the construct. This construct contains at least one oligo-T domain, but may also consist of further domains (see below). Proceeding from this double-stranded end, the reverse transcription is then effected, followed by the second strand synthesis. The resultant double-stranded product then has the domain structure
5′-biotin-PB/promoter-RAT-TES-cDNA-oligo-A/T-3′
and can be bound to a solid phase via the biotin group. If the TES is the recognition sequence of a type II restriction enzyme, the product can be cut therewith

GUARDANT HEALTH EX.1009.362

immediately. While the unbound portion of the cDNA can be washed away, a construct which remains bound on the solid phase has the domain sequence
5'-biotin-PB/promoter-RAT-TES-TAG-3',
where the TAG, which in this case has a defined length, characterizes the cDNA.

[0099] Alternatively, the TAG can also be obtained for the purposes of the invention by mechanical comminution of the cDNA or treatment with an unspecific endo- or exonuclease. In that case, however, it does not have a defined length by definition. In this case, there is no need to introduce a TES domain into the construct. The domain sequence
5'-biotin-PB/promoter-RAT-TAG
is sufficient to characterize the cDNA.

[0100] A further primer binding site (PB), an adapter or a marker may be bound to the 3' ends released after digestion or comminution. This construct, which has a defined length in the case of use of a tagging enzyme, can then be replicated by amplification proceeding from the two PBs present therein.

[0101] When the tagging enzyme is a type III endonuclease, for instance EcoP15I, preferably a construct containing a PB plus a TES is attached to the restriction site of the restriction enzyme or to the free end which arises from the unspecific endonuclease or the mechanical comminution and is in a distal position from the biotin group, so as to result in a construct of the
5'-biotin-PB/promoter-TAG-TES-PB-3'
form. In addition, it is also possible to insert a RAT here. In that case, the construct has the form:
5'-biotin-PB/promoter-TAG-TES-RAT-PB-3'.

GUARDANT HEALTH EX.1009.363

[0102] Constructs of this kind can be amplified by means of PCR, identified on the basis of their defined size and isolated (gel electrophoresis, HPLC).

[0103] In a particularly preferred embodiment of the production method, the solid phase used preferably comprises streptavidin-coated magnetic particles. Streptavidin-coated reaction vessels are also a preferred alternative. The particle-bound end of the nucleic acid strands produced should be protected against a ligation reaction.

2.3.) Constructs for the simultaneous quantification of 5′ and 3′ ends of cDNAs Above-described constructs of the form:

[0104] 5′-biotin-PB/T7 promoter-RAT-TES-cDNA-oligo-A/T-3′ or other constructs which have already been described, in principle, allow only one TAG to be obtained from one of the cDNA ends. In specific cases, however, the obtaining of two TAGs from the 5′ and 3′ end of the same cDNA is desired, which can thus be recognized as such. For this purpose, preference is given to producing a circular product of the
5′-PB-cDNA-oligo-A/T-PB′-3′
form where PB and PB′ preferably do not have the same sequence. Once PB and PB′ have been bound to the ends of the cDNA, the cDNA ends thus provided with known sequences can be transformed to a circular molecule by ligation of their PB ends under suitable conditions. These circular molecules can now be replicated by PCR proceeding from the PB domains. In order to be able to determine the original composition of the cDNA population with the aid of RATs, at least one end may be provided with a marker containing a PB domain. This can be effected via the CAP site and/or the poly-A end,

GUARDANT HEALTH EX.1009.364

as described above, in which case the circularization is preceded by formation of a construct of the
5'-PB-RAT-cDNA-3'
form at the 5' end of the cDNA following the above-described removal of the CAP site. At the 3' end, a construct of the
5'-cDNA-oligo-A/T-PB'-3' form or
5'-cDNA-oligo-A/T-RAT'-PB'-3' form is formed.

[0105] PB and PB' may have either complementary overhanging single strand ends which are particularly suitable for circularization or blunt ends. The ligation for circularization of the molecules is preferably effected in a sufficiently great dilution that the probability of one end of the same cDNA molecule being closer to the other end than any end of any other molecule is sufficiently great. Constructs containing a TAG from each end of the molecule can then be obtained from the molecules thus circularized either by enzymatic means or by mechanical comminution. They then have the following form:
5'-TAG-RAT-PB-PB'-RAT'-TAG-3'.

[0106] Attachment of two PBs to the ends of this construct gives rise to constructs of the following form:
1) 5'-PB''-TAG-RAT-PB-PB'-RAT-TAG-PB'''-3'
or, according to which 3' construct has been used,
2) 5'-PB''-TAG-RAT-PB-PB'-TAG-PB'''-3',
which can be amplified and then sequenced proceeding from PB'' and PB''', although PB'' and PB''' preferably do not necessarily have nonidentical sequences.

[0107] In order to obtain constructs of a defined length, a size selection may be effected, for example, by means of HPLC or gel electrophoresis, or it is possible to use one or more tagging enzymes.

[0108] For the use of type II enzymes, the 5′ end of cDNA is provided with a construct of the
5′-TES-(CAP site)-cDNA-3′
or
5′-PB-TES-(CAP site)-cDNA-3′
form, where the TES together with the PB is preferably part of an adapter.

3) Constructs for the quantification of cDNA which has not been produced from poly-A-containing RNA

[0109] Many RNA molecules in eukaryotic cells and virtually all bacterial and viral RNAs, unlike protein-encoding eukaryotic mRNAs, do not contain any 3′-poly-A sequence. In order to quantify them, they are preferably first ligated with a single- or double-stranded RNA oligonucleotide of known sequence, which can later serve as primer binding site. Single-stranded oligonucleotides can be ligated selectively to the 5′ or 3′ side of a single-stranded nucleic acid with the aid of single-strand RNA ligases, for example with T4 RNA ligase, by, for example, protecting one end of the oligonucleotide from ligation, while the 5′ end can be phosphorylated.

[0110] It is thus possible to selectively ligate an oligonucleotide containing a RAT and a PB (marker and adapter) onto either end of an RNA.

[0111] For example, it is possible to ligate RATs onto non-protein-encoding RNA and subsequently to transcribe them to DNA, and amplify and sequence them. Corresponding constructs have the following domain sequence:
5′-PB-RAT-RNA-PB-amino-C7-3′
or

GUARDANT HEALTH EX.1009.366

5'-PB-RNA-RAT-PB-amino-C7-3',
where all molecules are single-stranded, or PBs may also be present in the form of double-stranded RNA. As an alternative to ligation by means of RNA ligases, the RNA may be provided with a poly-A strand by synthetic means at the 3' end of the RNA with the aid of a poly(A) polymerase.

[0112] After insertion of the poly-A sequence and conversion to a double-stranded cDNA, it is also possible to quantify those RNAs having the same constructs as described above for the mRNAs. Since RNAs that do not encode proteins generally do not have any CAP structure, however, it is not possible to distinguish for the purposes of the method between full-length RNAs and fragmented molecules.

4) Constructs for the quantification of fragments of genomic DNA

4.1.) Constructs for the quantification of regions of genomic DNA: chromatin immunoprecipitation, digital karyotyping, methylation-specific digital karyotyping, metagenomics

[0113] A method for the quantitative determination of DNA-protein binding is based on a preparation of the sample by immunoprecipitation. Methods of this kind are frequently used for the determination of the binding sites for transcription factors. For example, genomic DNA which has not been freed of bound proteins is digested enzymatically with a restriction enzyme or an unspecifically cutting endonuclease, or comminuted in another way (for example by ultrasound). The DNA/protein complexes are precipitated by immunoprecipitation with an antibody against a particular protein (e.g. a transcription factor). The

DNA which is bound to the protein and is precipitated as well is freed of all proteins and converted to a form quantifiable by means of RATs with or without tagging enzyme. For this purpose, marker and optionally adapter are ligated to the ends of the precipitated DNA fragments, and the nucleic acid constructs present are amplified and subsequently sequenced. As a result, the amount of copies of a particular genomic DNA sequence, for example in a tissue sample, to which or close to which a protein was bound is obtained.

[0114] "Digital karyotyping" (DK) enables the quantification of short DNA sections in the genome. The technique serves for examination of chromosomal alterations, replications and deletions and for detection of the presence of foreign DNA. This involves first digesting the DNA to be examined with a (methylation-insensitive) restriction enzyme. It is then possible to ligate biotinylated markers or adapters. Thereafter, the sample is digested with a second, frequent-cutting (methylation-insensitive) restriction enzyme, and markers or adapters are ligated to the restriction sites that arise. The constructs which arise have to be provided with at least one marker and can thus be amplified and sequenced.

[0115] If markers or adapters have been equipped with TES, it is possible to use a tagging enzyme and, after binding to a solid phase, to purify either the bound or unbound marker-TAG or adapter-TAG combination and to provide it with a further marker or adapter, such that a marker together with an adapter (or a marker) in each case encloses a TAG. If markers and adapters have been provided with a TES, especially for EcoP15I, as envisaged in a preferred version, it is additionally possible to ligate bound and unbound marker-TAG and

GUARDANT HEALTH EX.1009.368

adapter-TAG combinations to one another, such that two TAGs are enclosed by the marker and/or adapter.

[0116] Methylation-specific digital karyotyping (MSDK): This technique serves to examine the methylation state of a genomic DNA. For this purpose, the first enzyme used in DK is replaced by a methylation-sensitive enzyme. This can cut only non-methylated recognition sequences. The rest of the steps are analogous to DK. According to the methylation state, different TAGs thus form from one sample.

[0117] There follows a description of a particularly preferred variant of DK, which includes the use of the tagging enzyme EcoP15I, in order to produce nucleic acid constructs for the quantitative determination of genomic DNA by the method of the invention.

[0118] For analysis of genomic DNA fragments by DK, for metagenomics applications or after immunoprecipitation, in a preferred method, the DNA, as described above, is first digested with one or more restriction enzymes. For the analysis of DNA originating from immunoprecipitated material, this can also be mechanically comminuted, for example, by shear or ultrasound.

[0119] Thereafter, the resultant DNA fragments are ligated with a biotinylated oligonucleotide 1 (adapter or marker) having a restriction sequence for a restriction enzyme in a distal position from the biotinylated end, giving rise to a ligation site (LS1) having an overhanging single-strand end after the restriction digestion with the appropriate enzyme. For ligation with mechanically comminuted DNA, oligonucleotides (markers or adapters) having blunt ends are used. The ends of the comminuted DNA are

GUARDANT HEALTH EX.1009.369

likewise blunted prior to ligation with the Klenow fragment. In addition, the adapter contains a primer binding site (PB1) at the biotinylated end. For the use of EcoP15I as tagging enzyme, the oligonucleotide 1 must additionally bear the sequence 5'-CAGCAG-3' (TES) upstream of the ligation site. After binding to a streptavidin-modified solid phase and washing for removal of unbound fragments, digestion is effected with a further enzyme. For this purpose, preference is given to using restriction enzymes having a maximum number of restriction sites in the DNA to be analyzed, for example NlaIII, DpnII, TaqI, FatI, Sau3A, MboI, etc. A further oligonucleotide (oligonucleotide 2) along with the bound DNA is ligated onto the new restriction site. The oligonucleotide 2 contains a further recognition sequence for the restriction enzyme EcoP15I (5'-CTGCTG-3' (TES)) followed by a restriction enzyme restriction site, which results in a ligation site (LS2) after a restriction digest. The ligation is effected with the DNA ligation sites which result from the digestion with the appropriate frequent-cutting restriction enzyme. Oligonucleotide 2 is additionally protected from ligation, for example by an amino-C7 modification, at the distal end from the LS.

[0120] The method steps described give rise to the following product:
5'-CTGCTG-LS2-nucleic acid-LS1-CAGCAG-PB1-3'-(solid phase).

[0121] Digestion with the tagging enzyme EcoP15I gives rise to TAGs of length 25-27 Bp. These can then be ligated to a further oligonucleotide 3. This oligonucleotide has the structure RAT-PB2, and ligation is effected only via the free end of the RAT, since the free end of PB2 is protected from ligation, for example by an amino-C7 modification. After washing the solid

phase, the bound nucleic acid constructs are detached from the solid phase, resulting in the following construct of particular suitability for high-throughput sequencing:

PB2-RAT-TAG-LS1-CAGCAG-PB1.

[0122] Alternatively or additionally, the RAT may also be integrated within the PB1-containing oligonucleotide 1, forming the product PB2-TAG-LS1-CAGCAG-RAT-PB1 or PB2-RAT-TAG-LS1-CAGCAG-RAT-PB1.

[0123] After the production of the second strand with a polymerase, digestion with an enzyme specific to the restriction site can produce a linker which has the base overhangs typical of the restriction site and hence enables ligation with a restriction site having complementary base overhangs. Alternatively, the ligation can also be effected with nucleic acid fragments having blunt restriction sites. In this way, in particular, direct linkage of a RAT with a TAG is possible without the intermediate connection of a linker. In principle, by introducing restriction sites via a linker sequence which serve for production of overhanging restriction sites, it is possible to achieve the incorporation of polynucleotides into the nucleic acid constructs in the desired orientation.

[0124] The degradation can be effected mechanically, for example by means of shear forces, physically, for example by means of ultrasound, or preferably by enzymatic degradation.

4.2.) Constructs for the quantification of both ends of fragments of genomic DNA with the aid of a circularization method

GUARDANT HEALTH EX.1009.371

[0125] As already mentioned, it is often desirable to obtain one TAG from each end of a nucleic acid molecule of a genomic DNA, in order to quantify both ends. For fragmentation of the genomic DNA, it can be divided into smaller fragments of any size either with restriction enzymes or unspecific endo- or exonucleases or by mechanical comminution. After binding of these linear molecules to the constructs already mentioned, having the form of (1) TES-RAT-PB-RAT-TES or (2) TES-RAT-PB-PB'-RAT-TES, the molecules may be circularized and then amplified if the fragments do not exceed a particular size.

[0126] These reactions can take place on a solid phase or preferably in solution. For binding to a solid phase, one option is the modification of one or both TES by a biotin group.

[0127] The amplification of the circular molecule proceeds from the primer binding sites (PB), where the primers in the case of (1) are at least partly complementary to one another and each become one strand of the PB. In the case of (2), there are two different primer binding sites and the amplification which proceeds from the primers that bind thereto proceeds in opposite directions. In both cases, the resultant circular molecules, after cutting with the TE, have the following domain sequence:

TAG-TES-RAT-PB-RAT-TAG

or

TAG-TES-RAT-PB-PB'-RAT-TAG.

[0128] It is often advisable (for example for directed sequencing or cloning) to bind one or two further domains consisting of one or two adapter(s), one or two linker(s) or one or two primer binding site(s) to one or both ends of these constructs such that the result

is then, for example, constructs having the domain sequence shown in s) to v):

cc) PB''-TAG-TES-RAT-PB-RAT-TAG

dd) PB''-TAG-TES-RAT-PB-RAT-TAG-PB''

ee) PB''-TAG-TES-RAT-PB-PB'-RAT-TAG-PB''

and

ff) PB''-TAG-TES-RAT-PB-PB'-RAT-TAG-PB'''.

[0129] In this case, the domains PB'' and PB''' here, in the context of the invention, may be replaced by the abovementioned domains (adapters).

Working examples:

Example 1: Use of RATs for the quantification of mRNAs

[0130] There follows a description by way of example of the use of the RATs for the quantification of mRNAs in a preferred method. For this purpose, the mRNA is first transcribed to a cDNA. Then a TAG provided with a marker containing a RAT is obtained from virtually every cDNA molecule. The resultant nucleic acid construct is then amplified and sequenced.

cDNA synthesis with an oligo-dT primer:

[0131] With the aid of a commercial kit comprising Trizol reagent (Invitrogen, Inc.), about 1 mg of total RNA is obtained from a biological sample (an MVV leukemia cell line). 5 µg of mRNA are isolated from the total RNA with a commercial kit ("Oligotex-Midi-Kit", Qiagen N.V.). The cDNA is subsequently produced with the "cDNA synthesis system" (Invitrogen Corp.), using the following oligonucleotide which has been biotinylated at the 5' end and contains the recognition sequence for EcoP15I CAGCAG:

GUARDANT HEALTH EX.1009.373

biotin-5'-CTGATCTAGAGGTACCGGATCCCAGCAGTTTTTTTTTTTTTTTT T-3'
(SEQ ID No. 2)

[0132] The product is then converted to a double-stranded DNA ("cDNA synthesis system", Invitrogen Corp.) and dissolved in 20 µL of LoTE buffer (3 mM Tris-HCl, pH 7.5, 0.2 mM EDTA).

Digestion with the anchoring enzyme DpnII:

[0133] The double-stranded cDNA (20 µL) is digested in a 200 µL reaction solution with 50 units of DpnII (New England BioLabs, Inc., Ipswich, UK; NEB) together with the buffer supplied at 37°C for 90 minutes. After the digestion, the cDNA is extracted to remove the enzyme with TE-equilibrated phenol/chloroform/isoamyl alcohol (25:24:1, pH = 8.0), precipitated with ethanol and dissolved in 100 µL of LoTE buffer.

[0134] 300 µL of streptavidin-coated magnetic particles (MP; Dynabeads M279, Dynal Biotech GmbH, Hamburg, DE) are washed with 200 µL of 1x B&W solution (5 mM Tris-HCl, pH 7.5, 0.5 mM EDTA, 1 M NaCl), and addition of the B&W solution is followed by utilization of the "Magnetic Particle Capturer" (MPC; Promega GmbH, Mannheim, DE) in order to keep the magnetic particles at the vessel wall and exchange the B&W solution. Added to the cDNA (100 µL) thereafter are 100 µL of 2 × B&W solution (10 mM Tris-HCl, pH 7.5, 1 mM EDTA, 2 M NaCl), and the 1x B&W solution of the MP is replaced by the cDNA solution. Thereafter, for binding of the biotinylated cDNA to the streptavidin-MPs, the MPs are incubated at room temperature under rotation for 30 minutes. With the aid of a magnet, the MPs to which the DpnII-digested cDNA has been bound are washed 2x with

200 µL of B&W solution and redissolved in 200 µL of LoTE.

Adapter ligation:

[0135] An adapter containing the recognition sequence for the tagging enzyme is ligated to the bound DNA. This adapter is composed of the following single-stranded oligonucleotides A and B: oligonucleotide B contains an amino-C7 modification at the 3′ end, in order to prevent unwanted ligation at this point, and has been phosphorylated at the 5′ end, in order to enable ligation on this side. The part of the 5′ end protected with the amino-C7 modification in the linker that will be double-stranded at a later stage serves as primer site for the later pyrosequencing reaction by the Solexa method.

[0136] For this purpose, the two individual strands of the oligonucleotide adapter

A: 5′-phosphate-GATCCTGCTGTAGAACTCTGAACCTGT [amino-C7]-3′ (SEQ ID No. 3)

and

B: 5′-ACAGGTTCAGAGTTCTACAGCAG-3′ (SEQ ID No. 4)

are dissolved in 0.2 mM Tris-EDTA in a concentration of 100 µM, and hybridized together to form a double strand by mixing the two oligonucleotide solutions in a ratio of 1:1, heating to 96°C and then gradually cooling to room temperature. The linker contains the recognition sequence for the tagging enzyme EcoP15I (CAGCAG). The result is a double-stranded linker having a 5′ GATC overhang which can be ligated to the DpnII restriction site.

GUARDANT HEALTH EX.1009.375

Ligase reaction:

[0137] The following ligation mixture is produced:
6 µL of 5 × ligase buffer (Invitrogen Corp.)
+ 5 µL of linker (100 µM)
+ 17 µL of H_2O
= 28 µL

[0138] The LoTE in the MP + cDNA mixture is replaced by the 28 µL of ligation mixture. The MPs are mixed cautiously, heated to 50°C for 2 minutes and cooled at room temperature for 15 minutes. 2 µL of T4 ligase (5 U/µL, Invitrogen Corp.) are added to the ligation mixture and the sample is incubated at 16°C for 2 hours; the sample is mixed cautiously every 20 minutes. After the ligation reaction, the unbound linkers are removed from the MPs by washing 3 times with 300 µL of 1 × B&W. The MPs are transferred into a new vessel and washed again 1x with 300 µL of 1 × B&W and then 2x with 1 × NEB4 buffer (New England Biolabs, Inc., NEB) and taken up in 300 µL of NEB4 buffer.

Cutting with the tagging enzyme:

[0139] The following reaction mixture is prepared:
10 µL of 10 × NEB3 buffer
10 µL of 10 mM ATP (NEB)
2 µL of EcoP15I (NEB)
1 µL of 100 × BSA (NEB)
77 µL of H_2O
= 100 µL

[0140] The 300 µL of NEB3 buffer in the MP + cDNA + linker mixture is replaced by the 100 µL of reaction mixture. The sample is incubated at 37°C for one hour with repeated cautious mixing. With the aid of the MPC,

the MPs are held at the vessel wall, while the supernatant can be removed and transferred into a new vessel. The supernatant contains the linker TAGs of length 52 bp (linker + TAG). The linker-TAG fragments in 100 µL of supernatant are extracted in a ratio of 1:1 with phenol/chloroform/isoamyl alcohol (25:24:1, pH = 8.0) solution, precipitated with 500 µL of ethanol + 67 µL of 7.5 M NH_4OAc + 2 µL of glycogen (NEB). After incubation at -20°C for 8 h, the sample is centrifuged at 4°C and 13 000 g for 30 minutes, washed twice with 70% ethanol, dried and dissolved in 20 µL of LoTE buffer.

Production of "blunted" adapter-TAG fragments:

[0141] Cutting with EcoP15I gives rise to adapter-TAG fragments with protruding 5′ ends. In order to ligate a RAT-containing marker having blunt ends thereto at a later stage, the ends are blunted. The blunting reaction takes place in 50 µL of 1 × NEB buffer 2 (New England Biolabs, Inc.) containing 33 µM dNTP, 5 units of polymerase I, large (Klenow) fragment (New England Biolabs, Inc.), at 25°C and with a 15-minute incubation time. Thereafter, the sample is made up to 200 µL with LoTE and extracted with phenol/chloroform/isoamyl alcohol (25:24:1, pH = 8.0) solution, precipitated with 1000 µL of ethanol + 133 µL of 7.5 M NH_4OAc + 2 µL of glycogen (NEB), and dissolved in 10 µL of LoTE.

Production and ligation with the RAT-containing marker:

The single-stranded marker having the following sequence:

[0142] phosphate 5′ NNNNNNNNTCGTATGCCGTCTTCTGCTTG 3′amino-C7 (=RAT-A), where $(N)_8$ is the single-stranded

RAT (SEQ ID No. 5), is incubated with the primer of the following sequence:

5′ CAAGCAGAAGACGGCATACGA-3′ (= RAT-B) (SEQ ID No. 6)

with the aid of the Klenow fragment in the following solution at 37°C for 30 minutes, and hence produced in double-stranded form:

5 µL of RAT A (10 µM)
5 µL of RAT B (10 µM)
1 µL of dNTPs (10 mM each)
5 µL of 10 × NEB buffer 2 (New England Biolabs, Inc.)
1 µL of Klenow fragment (New England Biolabs, Inc.)
+ 33 µL of H_2O
= 50 µL

[0143] The double-stranded marker containing the RAT is purified by means of phenol-chloroform, precipitated with ethanol, washed, dried and then taken up in 50 µL of H_2O.

[0144] Ligation with the adapter-TAG construct takes place under the following conditions:

10 µL of TAG solution in LoTE
1 µL of 0.1 mM marker
4 µL of 5 × ligation buffer (Invitrogen, Inc.)
2 µL of T4 ligase (Invitrogen, Inc.)
3 µL of H_2O
= 20 µL

[0145] The mixture is incubated at 16°C for 2 hours. Thereafter, the sample is made up to 200 µL with LoTE and extracted with phenol/chloroform/isoamyl alcohol (25:24:1, pH = 8.0) solution, precipitated with 1000 µL of ethanol + 133 µL of 7.5 M NH_4OAc + 2 µL of glycogen (NEB), and dissolved in 10 µL of LoTE.

PCR amplification:

[0146] First of all, various dilutions of the linker-TAG-RAT ligation mixture are used to conduct a PCR with 25 cycles in order to determine the concentration of best suitability for amplification. The PCR reaction mixtures, each in an amount of 20 µL, are incubated for the initial denaturing of the template DNA at 98°C for 2 minutes, followed by 25 cycles with: 98°C for 15 seconds, followed by 15 seconds at 60°C and finally 15 seconds at 72°C. The polymerase used is the Phusion polymerase (Finzymes, Finland).

[0147] The primers used were
5′ CAAGCAGAAGACGGCATACGA 3′ (SEQ ID No. 7)
and
5′ ACAGGTTCAGAGTTCTACAGCAG 3′ (SEQ ID No. 8).

[0148] The PCR products are applied to an 8% polyacrylamide gel. After staining with ethidium bromide and visualization under UV light, a 79-Bp marker-adapter-TAG band becomes visible in the gel. Thereafter, for preparative purposes, a 200 µL PCR mixture is run with that dilution which gave the best amplification.

Purification of the linker-TAG-RAT fragments:

[0149] The products from the preparative PCR are applied again to an 8% polyacrylamide gel and the piece of gel containing the 79 base pair (Bp) band is cut out with a scalpel. In a 1.5 mL Eppendorf vessel, the piece of gel is comminuted with a pestle and incubated in 500 µL of an elution solution (0.5% NH_4OAc, 2 mM EDTA, 0.1% SDS) at 37°C overnight. The eluted RAT-linker-TAG construct is then precipitated from the elution

solution with ethanol, washed 2× with 70% ethanol, dried and redissolved in 10 µL of LoTE.

[0150] The concentration and quality of the PCR product is determined by a gel electrophoresis. For the test, an aliquot of the purified RAT-linker-TAG construct is ligated into the pGEM®-T Easy vector (Promega), transformed into the *E. coli* strain DH5alpha, white colonies are selected and plasmids containing 5-10 RAT-linker-TAG constructs are sequenced by the Sanger method. Seven of the typical sequences thus obtained having different PB-RAT-TAG-LS-TES-PB' constructs are listed below. Each TAG is clearly marked by a RAT. The RAT domains of the constructs are in bold type, and the TAG domains in italic type.

Clone 1

CAAGCAGAAGACGGCATACGA**TCTCGACG***GCCACATCCTCGGTATGACAGGAGAT* *CCTGCTGTAGAACTCTGAA* (SEQ ID No. 9)

Clone 2

CAAGCAGAAGACGGCATACGA**GTCAAGCG***TGGGGGTGGGGGAGACACAATTTGAT* *CCTGCTGTAGAACTCTGAA* (SEQ ID No. 10)

Clone 3

CAAGCAGAAGACGGCATACGA**ACTTACAA***TTCTATGGTAGTACACACACACAGGA* *TCCTGCTGTAGAACTCTGAA* (SEQ ID No. 11)

Clone 4

CAAGCAGAAGACGGCATACGA**TTGATACC***TTCTCCAGGGCTCTGGTTACCTGATC* CTGCTGTAGAACTCTGAA (SEQ ID No. 12)

Clone 5

CAAGCAGAAGACGGCATACGA**CCCATCCC***CCCAAGGCTGATATTTCCCTGGGATC*
CTGCTGTAGAACTCTGAA (SEQ ID No. 13)

Clone 6

CAAGCAGAAGACGGCATACGA**TGTATTTG***ATTTATAATTTAATGGCTTGCAGATC*
CTGCTGTAGAACTCTGAACCTGT (SEQ ID No. 14)

Clone 7

CAAGCAGAAGACGGCATACGA**TGATGTAT***CCAATGGTTCCAGTCACATATTGATC*
CTGCTGTAGAACTCTGAACCTGT (SEQ ID No. 15)

[0151] With the aid of a BLAST search with the RAT-TAG construct in the NCBI GenBank, it was possible to assign each of the individual TAGs to a particular human gene (table 1):

Table 1: Tabular compilation of the results of the BLAST search

Clone no.	Result of the BLAST search
Clone 1	>gi|14993673|gb|AC010366.6| Homo sapiens chromosome 5 clone CTD-2044J15, complete sequence Length=115396 Score = 54.0 bits (27), Expect = 2e-05 Identities = 27/27 (100%), Gaps = 0/27 (0%) Strand=Plus/Plus Query 9 GCCACATCCTCGGTATGACAGGAGATC 35 ||||||||||||||||||||||||| Sbjct 6328 GCCACATCCTCGGTATGACAGGAGATC 6354
Clone 2	>gi|23272853|gb|BC035646.1| Homo sapiens hypothetical protein LOC284356, mRNA (cDNA clone IMAGE:5550805), with apparent retained intron Length=1330 Score = 56.0 bits (28), Expect = 5e-06 Identities = 28/28 (100%), Gaps = 0/28 (0%) Strand=Plus/Minus Query 8 GTGGGGGTGGGGGAGACACAATTTGATC 35 |||||||||||||||||||||||||| Sbjct 1285 GTGGGGGTGGGGGAGACACAATTTGATC 1258

Clone 3	>gi|10716633|gb|AC009404.5| Homo sapiens BAC clone RP11-28H22 from 2, complete sequence Length=112883 Score = 58.0 bits (29), Expect = 1e-06 Identities = 29/29 (100%), Gaps = 0/29 (0%) Strand=Plus/Plus Query 8 ATTCTATGGTAGTACACACACACAGGATC 36 ||||||||||||||||||||||||||| Sbjct 98244 ATTCTATGGTAGTACACACACACAGGATC 98272
Clone 4	>gi|142381010|ref|NM_002488.3| Homo sapiens NADH dehydrogenase (ubiquinone) 1 alpha subcomplex, 2, 8kDa (NDUFA2), mRNA Length=599 Score = 52.0 bits (26), Expect = 8e-05 Identities = 26/26 (100%), Gaps = 0/26 (0%) Strand=Plus/Minus Query 9 TTCTCCAGGGCTCTGGTTACCTGATC 34 ||||||||||||||||||||||||||| Sbjct 350 TTCTCCAGGGCTCTGGTTACCTGATC 325
Clone 5	>gi|146147624|gb|BC106894.2| Homo sapiens cDNA clone IMAGE:40030896 Length=673 Score = 52.0 bits (26), Expect = 8e-05 Identities = 26/26 (100%), Gaps = 0/26 (0%) Strand=Plus/Plus Query 9 CCCAAGGCTGATATTTCCCTGGGATC 34 ||||||||||||||||||||||||||| Sbjct 48 CCCAAGGCTGATATTTCCCTGGGATC 73
Clone 6	>gi|18376860|gb|AC011345.5| Homo sapiens chromosome 5 clone CTC-293E5, complete sequence Length=105797 Score = 44.1 bits (22), Expect = 0.018 Identities = 22/22 (100%), Gaps = 0/22 (0%) Strand=Plus/Plus Query 10 GCCATTAAATTATAAATCAAAT 31 |||||||||||||||||||||| Sbjct 27015 GCCATTAAATTATAAATCAAAT 27036
Clone 7	>gi|184692865|dbj|AK313066.1| Homo sapiens cDNA, FLJ93545 Length=1860 Score = 52.0 bits (26), Expect = 8e-05 Identities = 26/26 (100%), Gaps = 0/26 (0%) Strand=Plus/Plus Query 1 GATCAATATGTGACTGGAACCATTGG 26 ||||||||||||||||||||||||||| Sbjct 1555 GATCAATATGTGACTGGAACCATTGG 1580

There follows a sequence protocol in accordance with WIPO St. 25.

This can be downloaded from the official publication platform of the GPTMO.

CITATIONS PRESENT IN THE DESCRIPTION

This list of documents cited by the applicant has been generated automatically and is incorporated exclusively for better reader information. The list is not part of the German patent or utility model application. The GPTMO does not accept any responsibility whatsoever for any errors or omissions.

Patent literature cited

- US 5695937 [0006]
- US 6916633 [0085]

Non-patent literature cited

- S. M. Wang (2007) Understanding SAGE data, Trends in Genetics 23: 42–50 [0006]
- SAGE, V. E. Velculescu et al., Serial analysis of gene expression, Science 270 (1995), pp. 484–487 [0006]
- Nielsen KL, Høgh AL, Emmersen J., Nucleic Acids Res. 2006; 34(19): e133. DeepSAGE-digital transcriptomics with high sensitivity, simple experimental protocol and multiplexing of samples) [0006]
- S. Saha et al., Using the transcriptome to annotate the genome, Nat. Biotechnol. 20 (2002), pp. 508–512 [0006]
- Emmersen, J. et al., BMC Bioinformatics 2007, 8: 92 [0006]
- H. Matsumura et al., Gene expression analysis of plant host-pathogen interactions by SuperSAGE, Proc. Natl. Acad. Sci. U. S. A. 100 (2003), pp. 15718–15723 [0006]
- CAGE, T. Shiraki et al., Cap analysis gene expression for high-throughput analysis of transcriptional starting point and identification of promoter usage, Proc. Natl. Acad. Sci. U. S. A. 100 (2003), pp. 15776–15781) [0006]
- GIS, C. L. Wei et al., 5' Long serial analysis of gene expression (LongSAGE) and 3' LongSAGE for transcriptome characterization and genome annotation, Proc. Natl. Acad. Sci. U. S. A. 101 (2004), pp. 11701–11706 [0006]
- T. L. Wang et al., Digital karyotyping, Proc. Natl. Acad. Sci. U. S. A. 99 (2002), pp. 16156–16161 [0006]
- J. T. Park et al., Notch3 gene amplification in ovarian cancer, Cancer Res. 66 (2006), pp. 6312–6318 [0006]
- Min Hu et al., Methylation-specific digital karyotyping Nature Protocols 1, – 1621–1636 (2006) [0006]
- C. L. Wei et al., A global map of p53 transcription-factor binding sites in the human genome, Cell. 124 (2006), pp. 207–219 [0006]
- Krause et al., Phylogenetic classification of short environmental DNA fragments, Nucleic Acids Res. 2008 April; 36(7): 2230–2239 [0006]
- Torres T. T. et al., Gene expression profiling by massively parallel sequencing, Genome Res. 2008 18: 172–177 [0054]
- Dafforn et al., Biotechniques. 2004 Nov; 37 (5): 854–7 [0085]
- Caretti et al. 2008, J. Cellular Biochemistry 103: 556–563 [0085]
- Clepet et al. Improved fulllength cDNA production based on RNA tagging by T4 DNA-Ligase. Nucleic Acids Research, 2004, Vol. 32, No. 1 e6 [0094]

Claims:

1. A method for the quantitative determination of nucleic acids in a sample, **characterized in that**
a) nucleic acid constructs comprising
-at least one characterizing polynucleotide domain of a nucleic acid which is present in a sample and which is to be quantitatively determined, and
-at least one artificial polynucleotide domain with random sequence, and
-at least one primer binding site or polymerase binding site,
where one characterizing polynucleotide domain and at least one polynucleotide domain with random sequence form a characteristic domain combination in each case, which is flanked by at least one primer binding site or polymerase binding site,
are amplified in the presence of a polymerase, and
b) after the amplification, at least the sequence of the polynucleotide domains with random sequence and of a part of the characterizing polynucleotide domain are determined, it being possible, for the quantitative determination of the nucleic acid present in the sample, for combinations of identical sequence, or for combinations of identical sequence and combinations of similar sequence, of a characterizing polynucleotide domain and of a polynucleotide domain with random sequence to be identified as copies of a characterizing polynucleotide domain of the nucleic acid originally present in the sample.

2. The method for the quantitative determination of nucleic acids of claim 1, characterized in that the amplification is a PCR amplification or an amplification by cloning.

3. The method for the quantitative determination of nucleic acids of either of claims 1 and 2, characterized in that

a) a sample comprising a polynucleotide having a characterizing polynucleotide domain is admixed with a solution of a polynucleotide comprising an artificial polynucleotide domain with random sequence, at least one of the two polynucleotides possessing a primer binding site,

b) a ligation of at least one end of the polynucleotides comprising the characterizing polynucleotide domains with the polynucleotide comprising the polynucleotide domain with random sequence, so that the primer binding site is not located between the characterizing polynucleotide domain and the polynucleotide domain with random sequence, and

c) the resulting nucleic acid constructs, marked with a polynucleotide domain with random sequence, are multiplied by amplification with a polymerase, and at least the random sequence and the sequence of the characterizing polynucleotide domains are ascertained.

4. The method of claim 3, characterized in that an end of the polynucleotides comprising a characterizing polynucleotide domain and/or an end of the polynucleotides comprising a polynucleotide domain with random sequence are protected from ligation and/or enzymatic digestion, and, if a primer binding site is present in the polynucleotide, this end is protected from ligation and/or enzymatic degradation.

5. The method of claim 4, characterized in that the polynucleotides comprising a characterizing polynucleotide domain and protected at one end against restriction digestion are digested with a restriction enzyme, before the polynucleotides comprising a

GUARDANT HEALTH EX.1009.386

polynucleotide domain with random sequence are ligated onto the free end of the remaining protected polynucleotide fragment.

6. The method of any of claims 3 to 5, characterized in that the polynucleotides comprise a binding site for the attachment of a tagging enzyme and, after ligation of the polynucleotides comprising the characterizing polynucleotide domains with the polynucleotides comprising a polynucleotide domain with random sequence, and after attachment of the tagging enzyme from the ligated polynucleotides obtained, the tagging enzyme is used to eliminate nucleic acid constructs which comprise a characterizing polynucleotide domain and which comprise a polynucleotide domain with random sequence.

7. The method of claim 6, characterized in that the tagging enzyme used is a type II or type III restriction enzyme.

8. The method of either of claims 6 and 7, characterized in that the tagging enzyme used is EcoP15I, MmeI or BsmFI.

9. The method of claim 1, characterized in that the nucleic acid construct used has one of the following domain sequences:
PB~TAG~RAT,
PB~RAT~TAG,
PB~TAG~RAT~PB',
PB~RAT~TAG~RAT,
PB~RAT~TAG~RAT~PB',
PB~RAT~TAG~TAG~RAT,
PB~RAT~$(TAG)_n$~PB; where n = 2 to 50, preferably where n = 2 to 10,

GUARDANT HEALTH EX.1009.387

PB-RAT-$(TAG)_n$-RAT-PB; where n = 2 to 50, preferably where n = 2 to 10,

PB-RAT-$(TAG)_n$; where n = 2 to 50, preferably where n = 2 to 10,

PB-RAT-TAG-RAT-TAG,

PB-TAG-RAT-TAG-RAT,

PB-RAT-TAG-RAT-TAG-PB',

PB-$(RAT-TAG)_n$-PB'; where n = 3 to 50, preferably where n = 3 to 10,

PB-RAT-TAG-PB'-PB''-RAT-TAG-PB''',

PB-RAT-$(TES)_m$-TAG-$(TES)_m$-PB'-PB''-RAT-$(TES)_m$-TAG-$(TES)_m$-PB'''-PB''''-RAT-$(TES)_m$-TAG-$(TES)_m$-PB'''''; where m = 0 or 1 for each m independently of one another, and at least one m = 1,

PB-RAT-$(TES)_m$-TAG-$(TES)_m$-PB'-(PB-RAT-$(TES)_m$-TAG-$(TES)_m$--PB-$(TES)_m$-TAG-$(TES)_m$-RAT-PB' where n = 0 to 10 and where m = 0 or 1 for each m independently of one another, and at least one m = 1,

TAG-TES-RAT-PB-RAT-TES,

TAG-TES-RAT-PB-PB'-RAT-TES,

PB''-TAG-TES-RAT-PB-RAT-TAG,

PB''-TAG-TES-RAT-PB-RAT-TAG-PB',

PB''-TAG-TES-RAT-PB-PB'-RAT-TAG-PB''',

TAG-RAT-PB-RAT,

TAG-RAT-PB-PB'-RAT,

PB''-TAG-RAT-PB-RAT-TAG,

PB''-TAG-RAT-PB-RAT-TAG-PB',

PB''-TAG-RAT-PB-PB'-RAT-TAG-PB''',

PB-TAG-RAT-TAG, or

PB-TAG-RAT-TAG-PB', where

PB, PB', PB'', PB''', PB'''', PB''''' are primer binding sites which may possess identical or different sequences, RAT is a polynucleotide domain with random sequence, TAG is a polynucleotide domain with characterizing sequence, and TES is a tagging enzyme binding site.

GUARDANT HEALTH EX.1009.388

10. The method of claim 1, characterized in that the nucleic acid construct used has one of the following domain sequences:
cDNA-oligo-A/T-RAT-promoter",
cDNA-oligo-A/T-RAT-PB-promoter",
cDNA-oligo-A/T-TES-RAT-PB,
5'-promoter-RAT-mRNA-3',
5'-biotin-PB/promoter-RAT-TES-mRNA,
5'-biotin-PB/promoter-RAT-TES-cDNA-oligo-A/T-3',
5'-biotin-PB/promoter-RAT-TES-TAG-3',
5'-biotin-PB/promoter-RAT-TAG,
5'-biotin-PB/promoter-TAG-TES-PB-3',
5'-biotin-PB/promoter-TAG-TES-RAT-PB-3',
5'-PB-cDNA-oligo-A/T-PB'-3',
5'-PB-RAT-cDNA-3',
5'-cDNA-oligo-A/T-PB'-3',
5'-cDNA-oligo-A/T-RAT'-PB'-3',
5'-TAG-RAT-PB-PB'-RAT'-TAG-3',
5'-PB''-TAG-RAT-PB-PB'-RAT-TAG-PB'''-3',
5'-PB''-TAG-RAT-PB-PB'-TAG-PB'''-3',
5'-TES-cDNA-3',
5'-PB-TES-cDNA-3',
5'-PB-RAT-RNA-PB-amino-C7-3',
5'-PB-RNA-RAT-PB-amino-C7-3',
5'-PB-RAT-RNA-PB-amino-C7-3',
5'-PB-RNA-RAT-PB-amino-C7-3',
TAG-TES-RAT-PB-RAT-TAG, or
TAG-TES-RAT-PB-PB'-RAT-TAG.

11. A marker ensemble for the quantitative determination of nucleic acids, characterized in that the markers contained comprise an artificial polynucleotide domain each with random sequences different from one another, and a primer binding site.

12. The marker ensemble of claim 11, characterized in that the markers comprise a binding site for a tagging

GUARDANT HEALTH EX.1009.389

enzyme, the tagging enzyme binding site being located at the distal end relative to the primer binding site of the marker.

13. The use of an ensemble of markers of either of claims 11 and 12 for the quantitative determination of nucleic acids in a sample.

14. The use of a method of any of claims 1 to 8 for the quantitative determination of mRNA, cDNA, aptamer DNA, or genomic DNA, for the methylation-specific quantitative determination of genomic DNA, for the quantitative determination of protein-bound genomic DNA.

There follow 4 pages of drawings

Appended drawings

Fig. 1

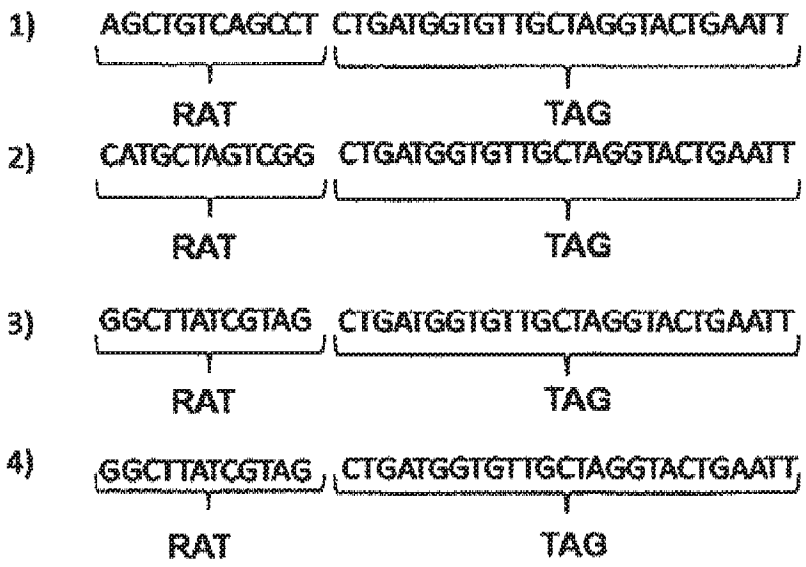


Fig. 2

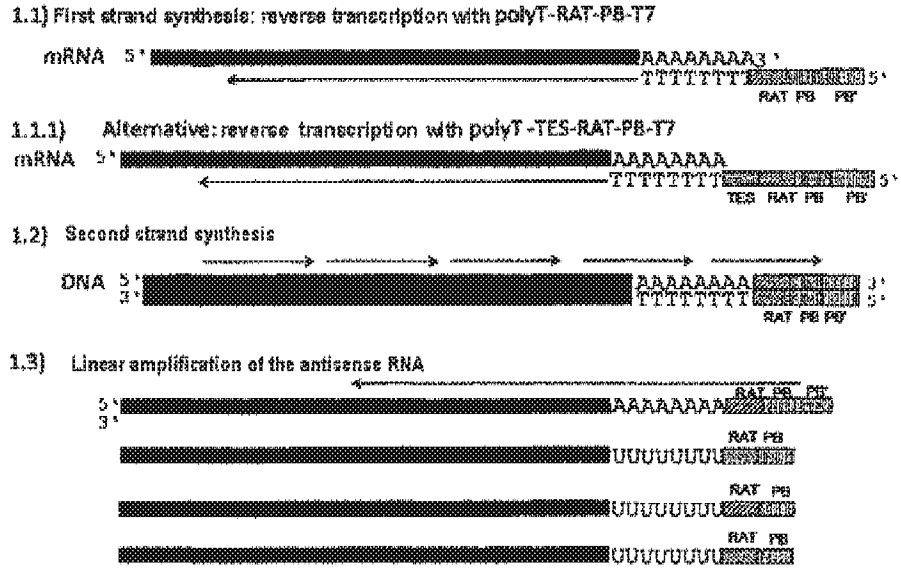


Fig. 2 (Continued)

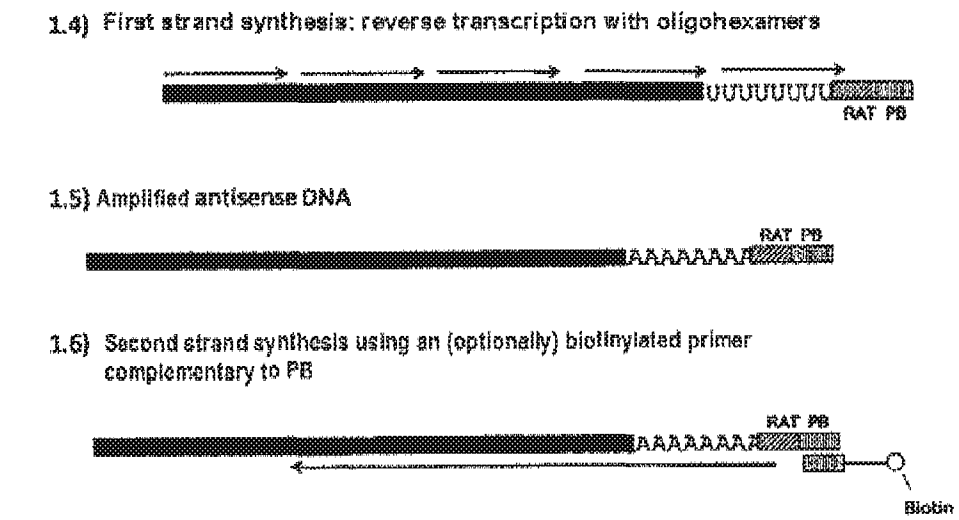


Fig. 2 (Continued)

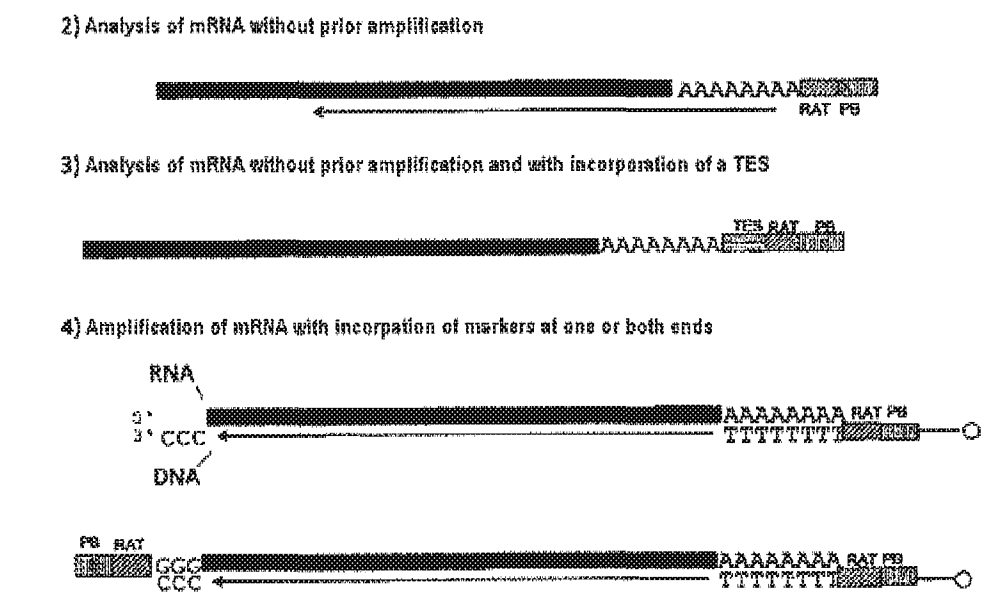


Fig. 3

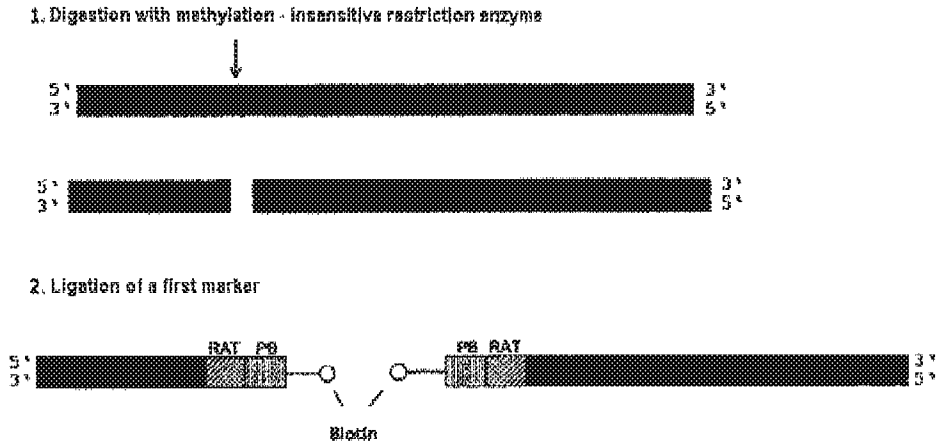


Fig. 3 (continued)

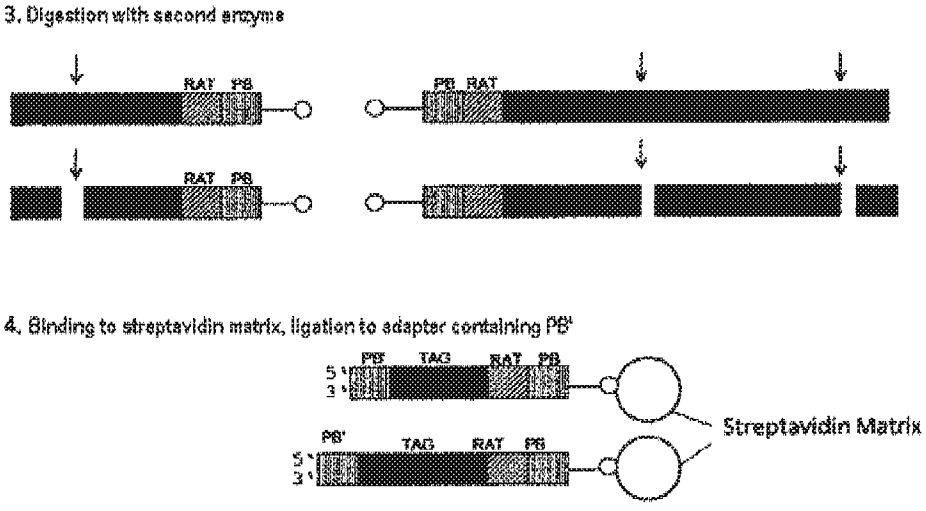
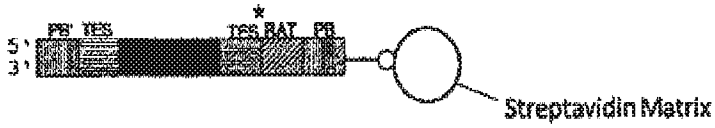


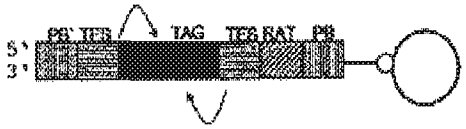
Fig. 4

5) Nucleic acid constructs formed with marker and adapter containing a tagging enzyme binding site (TES) in a distal position relative to the PB or PB' replaces step 4 from fig. 3



*TES optional for particular tagging enzymes (MmeI, BsmFI) but obligatory for EcoP15I.

6) Digestion with tagging enzyme



7.1) Use of the unbound end as a tag:



7.2) Use of the bound end as a tag:

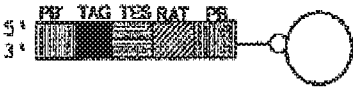


Fig. 4 (continued)

7.3) Use of both ends as a tag



8.) Sequencing

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Miscellaneous:				
SUBMISSION- INFORMATION DISCLOSURE STMT	2806	1	120	120
		Total in USD ($)		**120**

Electronic Acknowledgement Receipt

EFS ID:	35246258
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	25-FEB-2019
Filing Date:	07-MAR-2016
Time Stamp:	19:41:49
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$120
RAM confirmation Number	022619INTEFSW00023841033125
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.397

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		190225_81503-AA_IDS_ADR.pdf	173861 1de4d78711108af271cc47c149bd9a870900e647	yes	5
	Multipart Description/PDF files in .zip description				
	Document Description		**Start**	**End**	
	Transmittal Letter		1	2	
	Patent Term Adjustment Stmt Under 37CFR1.704(d)		3	3	
	Transmittal Letter		4	5	
Warnings:					
Information:					
2	Information Disclosure Statement (IDS) Form (SB08)	190225_81503-AA_Exhibit_A_ADR.pdf	315154 3da27a4be8b819ac1d12d8fda938402ddde f4712	no	1
Warnings:					
Information:					
This is not an USPTO supplied IDS fillable form					
3	Non Patent Literature	190225_81503-AA_Exhibit_1_ADR.pdf	145342 84f80c296e4acdeeaca22b440056aba51bd85af4	no	1
Warnings:					
Information:					
4	Non Patent Literature	190225_81503-AA_Exhibit_2_ADR.pdf	2720941 33430f1d7f8c70b5e3eedd27b170d6e6717e2f6e	no	17
Warnings:					
Information:					

5	Foreign Reference	190225_81503-AA_Exhibit_3_ADR.pdf	3529919 5df2ca5b656cb2f8dd40e8b0b457d333a094f521	no	76
Warnings:					
Information:					
6	Non Patent Literature	190225_81503-AA_Exhibit_4_ADR.pdf	2495684 d797dbdfeb63ed1492622eb304162fdc31ea4f71	no	8
Warnings:					
Information:					
7	Non Patent Literature	190225_81503-AA_Exhibit_5_ADR.pdf	3024110 2ca9f32e10b79c742689e53cb1e2d67ecc3374f8	no	10
Warnings:					
Information:					
8	Non Patent Literature	190225_81503-AA_Exhibit_6_ADR.pdf	7968861 37af69188b5dc19b06d697af51f1a0958a411656	no	12
Warnings:					
Information:					
9	Fee Worksheet (SB06)	fee-info.pdf	30424 66828c81102cd70798f88c0d3b548bf72a12b81a	no	2
Warnings:					
Information:					
			Total Files Size (in bytes):	20404296	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s) : James Hicks et al.

U.S. Serial No. : 15/063,278 Examiner: Frank W. M. Lu

Filing Date : March 7, 2016 Art Unit: 1634

Conf. No. : 4038

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

30 Rockefeller Plaza
20th Floor
New York, New York 10112
February 25, 2019

BY EFS
Commissioner for Patents
Alexandria, VA

FOURTH INFORMATION DISCLOSURE STATEMENT

In accordance with their duty of disclosure under 37 C.F.R. § 1.56 and §1.97(a)-(d), applicants request that the documents listed hereunder and on the substitute Form-1449, attached hereto as **Exhibit A**, be considered and made of record in the above-identified application.

This Fourth Information Disclosure Statement lists documents arising out of a Notice of opposition to a European Patent filed on January 25, 2019 against counterpart foreign patent, European Patent No. EP2630263, cited in a Communication of a notice of opposition issued by the European Patent Office on February 1, 2019.

Reference item **1** is a Communication of a notice of opposition issued by the European Patent Office on February 1, 2019 in connection with EP2630263. Reference item **2** is the Notice of opposition to a European Patent filed January 25, 2019 against counterpart foreign patent, European Patent No. EP2630263 cited in reference item **1**. Each of reference items **3-8** were cited in reference item **2**.

Reference items **3** and **4** are U.S. Patent application publications. Pursuant to 37 C.F.R 1.98(a)(2)(ii), copies of references **3** and **4** are not being submitted.

Copies of reference items **1, 2,** and **5-8** are attached hereto as **Exhibits 1-6,** respectively.

1. Communication of a notice of opposition, issued by the European Patent Office on February 1, 2019, concerning European Patent No. EP2630263 (**Exhibit 1**);

2. Notice of opposition to a European Patent, filed January 25, 2019, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263 (**Exhibit 2**);

3. U.S. Patent Application Publication No. 2008-0261204, published October 23, 2008 (Lexow)

4. U.S. Patent Application Publication No. 2007-0020640, published January 25, 2007 (McCloskey et al.)

5. English Translation of German Patent Publication No. DE 10 2008 025 656 A1, published December 3, 2009 (Björn Rotter) (**Exhibit 3**);

6. Chiang et al. (2008) "High-resolution Mapping of Copy-number Alterations with Massively Parallel Sequencing", Nature Methods 6(1):99-103 (**Exhibit 4**);

7. Hug and Schuler (2003) "Measurement of the Number of Molecules of a Single mRNA Species in a Complex mRNA Preparation", J. theor. Biol. 221:615-624 (**Exhibit 5**); and

8. Meyerson et al. (2010) "Advances in Understanding Cancer Genomes Through Second-Generation Sequencing", Nature Reviews 11:685-696 (**Exhibit 6**).

This Information Disclosure Statement is being submitted under 37 C.F.R. §1.97(d), *viz.* after the mailing of a final action under 37 C.F.R.

§1.113, a notice of allowance under 37 C.F.R. §1.311, or an action that closes prosecution in the application and before the payment of the issue fee.

In accordance with 37 C.F.R. §1.97(d)-(e) the undersigned certifies that reference item **2** contained in this Information Disclosure Statement was first cited in a communication from a foreign patent office in a counterpart foreign application not more than three months prior to the filing of the information disclosure statement, i.e. cited in reference item **1**.

Additionally, in accordance with 37 C.F.R. §1.704(d)(ii), reference item **1** is a communication that was issued by a patent office in a counterpart foreign or international application or by the Office, and this communication was not received by any individual designated in §1.56(c) more than thirty days prior to the filing of the information disclosure statement. In accordance with 37 C.F.R. §1.704(d)(i), reference item **2** was first cited in reference item **1**, a communication from a patent office in a counterpart foreign or international application or from the Office, and this communication was not received by any individual designated in §1.56(c) more than thirty days prior to the filing of this information disclosure statement.

Accordingly, per 37 C.F.R. §1.704(d), this paper containing only an information disclosure statement in compliance with 37 C.F.R. §§1.97-1.98 will not be considered a failure to engage in reasonable efforts to conclude prosecution of this application under 37 C.F.R §§1.704(c)(6), (c)(6), (c)(8), (c)(9), or (c)(10), and will thus not result in a reduction of any potential adjustment of the term of any patent issuing from this application under 37 C.F.R. §1.703(a)-(e).

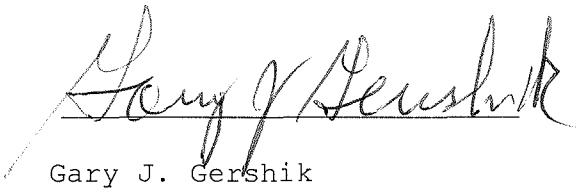
Per 37 C.F.R. §1.97(d)(2) and 37 C.F.R. §1.17(p), the fee for submitting this Fourth Supplemental Information Disclosure Statement is TWO HUNDRED FORTY DOLLARS ($240.00), and the undersigned hereby authorizes charging this amount to Deposit Account No. 03-3125.

The Examiner is respectfully requested to make of record each item listed on the substitute Form PTO-1449 by initialing and dating the attached substitute Form PTO-1449, and returning a copy of the initialed and dated form to applicants' undersigned attorney.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

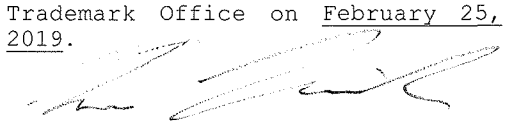
No fee, other than the $240.00 fee for which authorization to charge Deposit Account No. 03-3125 was given hereinabove, is deemed necessary in connection with the filing of this Fourth Information Disclosure Statement. If any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,



Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on February 25, 2019.



Aditya Ramanathan

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☑ SMALL ☐ MICRO

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A		N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A		N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*		x $40 =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*		x $210 =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).				
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))					
* If the difference in column 1 is less than zero, enter "0" in column 2.				TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT	04/17/2019	CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0		x $50 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	*** 3	= 0		x $230 =	0
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	0

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT		CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=		x $0 =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=		x $0 =	
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.	SLIE
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".	/HELENA L PAYTON/
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".	
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.	

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

Doc Code: A.NE.AFCP
Document Description: After Final Consideration Pilot Program Request

PTO/SB/434 (05-13)

CERTIFICATION AND REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0

Practitioner Docket No.:	Application No.:	Filing Date:
5981/81503-AA	15/063,278	3/7/2016
First Named Inventor:	**Title:**	
James Hicks	Varietal Counting of Nucleic Acids for Obtaining Genomic Copy Number Information	

APPLICANT HEREBY CERTIFIES THE FOLLOWING AND REQUESTS CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0 (AFCP 2.0) OF THE ACCOMPANYING RESPONSE UNDER 37 CFR 1.116.

1. The above-identified application is (i) an original utility, plant, or design nonprovisional application filed under 35 U.S.C. 111(a) [a continuing application (*e.g.*, a continuation or divisional application) is filed under 35 U.S.C. 111(a) and is eligible under (i)], or (ii) an international application that has entered the national stage in compliance with 35 U.S.C. 371(c).
2. The above-identified application contains an outstanding final rejection.
3. Submitted herewith is a response under 37 CFR 1.116 to the outstanding final rejection. The response includes an amendment to at least one independent claim, and the amendment does not broaden the scope of the independent claim in any aspect.
4. This certification and request for consideration under AFCP 2.0 is the only AFCP 2.0 certification and request filed in response to the outstanding final rejection.
5. Applicant is willing and available to participate in any interview requested by the examiner concerning the present response.
6. This certification and request is being filed electronically using the Office's electronic filing system (EFS-Web).
7. Any fees that would be necessary consistent with current practice concerning responses after final rejection under 37 CFR 1.116, *e.g.*, extension of time fees, are being concurrently filed herewith. [There is no additional fee required to request consideration under AFCP 2.0.]
8. By filing this certification and request, applicant acknowledges the following:
    - Reissue applications and reexamination proceedings are not eligible to participate in AFCP 2.0.
    - The examiner will verify that the AFCP 2.0 submission is compliant, *i.e.*, that the requirements of the program have been met (see items 1 to 7 above). For compliant submissions:
        - The examiner will review the response under 37 CFR 1.116 to determine if additional search and/or consideration (i) is necessitated by the amendment and (ii) could be completed within the time allotted under AFCP 2.0. If additional search and/or consideration is required but cannot be completed within the allotted time, the examiner will process the submission consistent with current practice concerning responses after final rejection under 37 CFR 1.116, *e.g.*, by mailing an advisory action.
        - If the examiner determines that the amendment does not necessitate additional search and/or consideration, or if the examiner determines that additional search and/or consideration is required and could be completed within the allotted time, then the examiner will consider whether the amendment places the application in condition for allowance (after completing the additional search and/or consideration, if required). If the examiner determines that the amendment does not place the application in condition for allowance, then the examiner will contact the applicant and request an interview.
            - The interview will be conducted by the examiner, and if the examiner does not have negotiation authority, a primary examiner and/or supervisory patent examiner will also participate.
            - If the applicant declines the interview, or if the interview cannot be scheduled within ten (10) calendar days from the date that the examiner first contacts the applicant, then the examiner will proceed consistent with current practice concerning responses after final rejection under 37 CFR 1.116.

Signature	Date
Gary J. Gershik	April 17, 2019
Name (Print/Typed)	**Practitioner Registration No.**
Gary J. Gershik	39,992

Note: *This form must be signed in accordance with 37 CFR 1.33. See 37 CFR 1.4(d) for signature requirements and certifications. Submit multiple forms if more than one signature is required, see below*.*

☐ * Total of _____ forms are submitted.

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
April 17, 2019

BY EFS
Commissioner for Patents
Alexandria, VA

AMENDMENT IN RESPONSE TO NOVEMBER 30, 2018 FINAL OFFICE ACTION AS A REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0 AND PETITION FOR A TWO-MONTH EXTENSION OF TIME

This Amendment is in response to the November 30, 2018 Final Office Action issued by the United States Patent and Trademark Office in connection with the above-identified application. A response to the November 30, 2018 Final Office Action was originally due February 28, 2019. Applicants hereby petition for a two-month extension of time. The required fee for a two-month extension of time is THREE HUNDRED DOLLARS ($300.00) for a small entity and authorization is hereby given to charge this fee to Deposit Account No. 03-3125. With a two-month extension of time, a response to the November 30, 2018 Final Office Action is now due April 30, 2019. Accordingly, this Amendment is being timely filed.

Amendments to the Drawings are on page **3** of this paper.

Amendments to the Claims begin on page **4** of this paper.

Remarks begin on page **12** of this paper.

A **Terminal Disclaimer** is referred to on page **19** of this paper and is attached hereto as **Exhibit 3.**

A **Request to Update the Name of the Applicant** begins on page **21** of this paper.

AMENDMENTS TO THE DRAWINGS

Applicants attach hereto as **Exhibit 1** EIGHT (8) replacement sheets of drawings containing Figures 7-10.

AMENDMENTS TO THE CLAIMS

Pursuant to 37 C.F.R. § 1.121(c), this listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

1-67. (Canceled)

68. (Currently amended) A method for obtaining genomic copy number information from genomic nucleic acids, comprising:

a) obtaining segments of the genomic nucleic acids;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each of the tag associated sequence reads of step (d); and

f) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome thereby obtaining a count for each location on the genome, wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome,

thereby obtaining genomic copy number information from the genomic nucleic acids.

69. (Currently amended) The method of claim 68, further comprising estimating a genomic copy number of multiple locations ~~on the genome~~ within a region of the genome.

70. (Previously presented) The method of claim 68, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative genomic copy number of the location.

71. (Previously presented) The method of claim 68, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value

obtained in step (h),

thereby estimating the relative genomic copy number of the first region of the genome to the genomic copy number of the second region of the genome.

72. (Previously presented) The method of claim 71, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (h) and the counts obtained in step (j).

73. (Previously presented) The method of claim 71, wherein the second region of the genome comprises a centromere.

74. (Previously presented) The method of claim 68, further comprising (l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative genomic copy number of the region of the genome.

75. (Currently amended) A method for obtaining mRNA copy number information from mRNA transcripts, comprising:

a) Generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) in the amplified tagged nucleic acid molecules, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome,

thereby obtaining mRNA copy number information from the mRNA transcripts.

76. (Withdrawn - Currently amended) The method of claim 68, wherein said tagging the segments comprises:

n) adding a polynucleotide tail to the ends of the segments of the genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail;

o) subjecting the zero-th order derivative strands of step (n) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands under conditions that promote the formation of only one complement of the zero-th order derivative strands, thereby generating first order derivative strands;

p) adding a polynucleotide tail to the first order derivative strands, thereby generating a first order derivative strands comprising the polynucleotide tail;

q) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the first order derivative strands under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps o) and q) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the unique tagged nucleic acid molecules.

77. (Currently amended) The method of claim 68, wherein said tagging the segments comprises:

r) adding a polynucleotide tail to the ends of the segments of genomic nucleic acids, thereby generating zero-th order derivative strands comprising the polynucleotide tail,

s) subjecting the zero-th order derivative strands of step (r) to a ligation reaction in the presence of primers capable of hybridizing to the polynucleotide tail of the zero-th order strands added in step (r) under conditions that promote the ligation of a primer to the 5′ ends of the zero-th order derivative strands,

t) subjecting the product of step (s) to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step r) under conditions which promote the formation of only one complement of the product of step (s), wherein the primers of step (s) and the

primers of step (t) have different nucleotide sequences, and wherein the polymerase has 3'-5' proofreading activity,

wherein the primers of at least one of steps (s) and (t) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, thereby generating the unique tagged nucleic acid molecules.

78. (Previously presented) The method of claim 77, wherein said adding the polynucleotide tail comprises the use of a terminal transferase.

79. (Previously presented) The method of claim 68, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to the PCR or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to step (d).

80. (Previously presented) The method of claim 68, wherein the tags further comprise a sample tag.

81. (Currently amended) The method of claim 80, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules ~~product~~ of step (c).

82. (Previously presented) The method of claim 81, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

83. (Previously presented) The method of claim 68, wherein the unique tagged nucleic acid molecules are generated from a single cell.

84. (Previously presented) The method of claim 79, wherein the unique tagged nucleic acid molecules are subjected to hybrid capture prior to the PCR in step (c).

85. (Currently amended) The method of claim 84, wherein each of the unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array.

86. (Previously presented) The method of claim 79, wherein the amplified unique tagged nucleic acid molecules produced in step (c) are subjected to hybrid capture prior to said sequencing the amplified unique tagged nucleic acid molecules in step (d).

87. (Currently amended) The method of claim 86, wherein each of the amplified unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on a microarray.

REQUEST TO UPDATE THE NAME OF THE APPLICANT PURSUANT TO 37 C.F.R. § 1.46(C)

Applicant has attached hereto as **Exhibit 3**, a Terminal Disclaimer signed by an authorized official of the assignee of record of the subject application, namely Cold Spring Harbor Laboratory. Applicant understands the assignee must be named applicant or must become applicant in order for the Terminal Disclaimer to be accepted.

Accordingly, applicant hereby requests under 37 C.F.R. § 1.46(c) that the applicant of the subject application be changed to Cold Spring Harbor Laboratory.

37 C.F.R. § 1.46(c) requires that a request to change the applicant include an Application Data Sheet under 37 C.F.R. § 1.76 specifying the applicant in the applicant information section and comply with 37 C.F.R. §§ 3.71 and 3.73.

Accordingly, a corrected Application Data Sheet (ADS) identifying Cold Spring Harbor Laboratory as the applicant of the subject application is attached hereto as **Exhibit 5**.

The corrected ADS identifies the information being changed, with underlining for insertions and strike-through for text removed, in compliance with 37 C.F.R. § 1.76(c)(2). The Terminal Disclaimer establishes that Cold Spring Harbor Laboratory is the assignee of the entire right, title, and interest in the above-identified application pursuant to 37 C.F.R. § 3.73(c). Additionally, applicant attaches hereto as **Exhibit 4** a statement under 37. C.F.R. §3.73(c). Both the Statement under 37 C.F.R. §3.73(c) and the Terminal Disclaimer establish that Cold Spring Harbor Laboratory is the assignee of the entire right, title, and interest in the above-identified application pursuant to 37 C.F.R. §3.73(c). As assignee of the above-identified application, Cold Spring Harbor Laboratory is entitled to conduct

prosecution of the subject application as applicant pursuant to 37 C.F.R. § 3.71.

Additionally, applicant attaches hereto as **Exhibit 6** a Power of Attorney and Statement under 37 C.F.R. § 3.73(b) signed by authorized officials of Cold Spring Harbor Laboratory. Both the Power of Attorney and Statement under 37 C.F.R. § 3.73(b) and the Terminal Disclaimer establish that Cold Spring Harbor Laboratory is the assignee of the entire right, title, and interest in the above-identified application pursuant to 37 C.F.R. § 3.73(c).

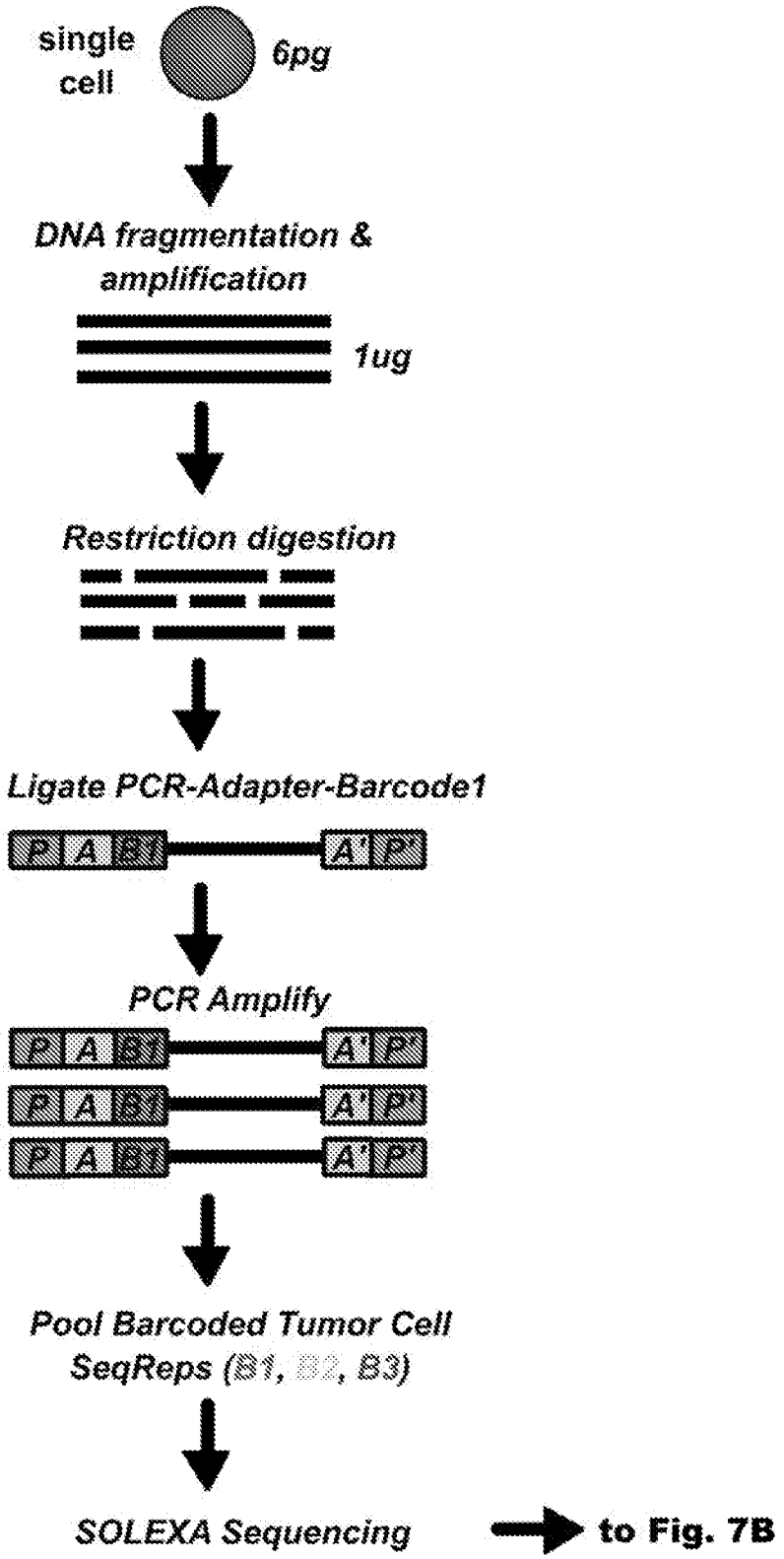
Applicant respectfully requests that the United States Patent and Trademark Office

i) issue a corrected filing receipt identifying "Cold Spring Harbor Laboratory" as applicant; and

ii) approve the Terminal Disclaimer attached hereto as **Exhibit 3**,

upon the granting of this Request.

Fig. 7A



U.S. Serial No.: 15/063,278
Applicant(s): James Hicks et al.
Filed: March 7, 2016
Exhibit 1

GUARDANT HEALTH EX.1009.422

Fig. 7B (continued from Fig. 7A)

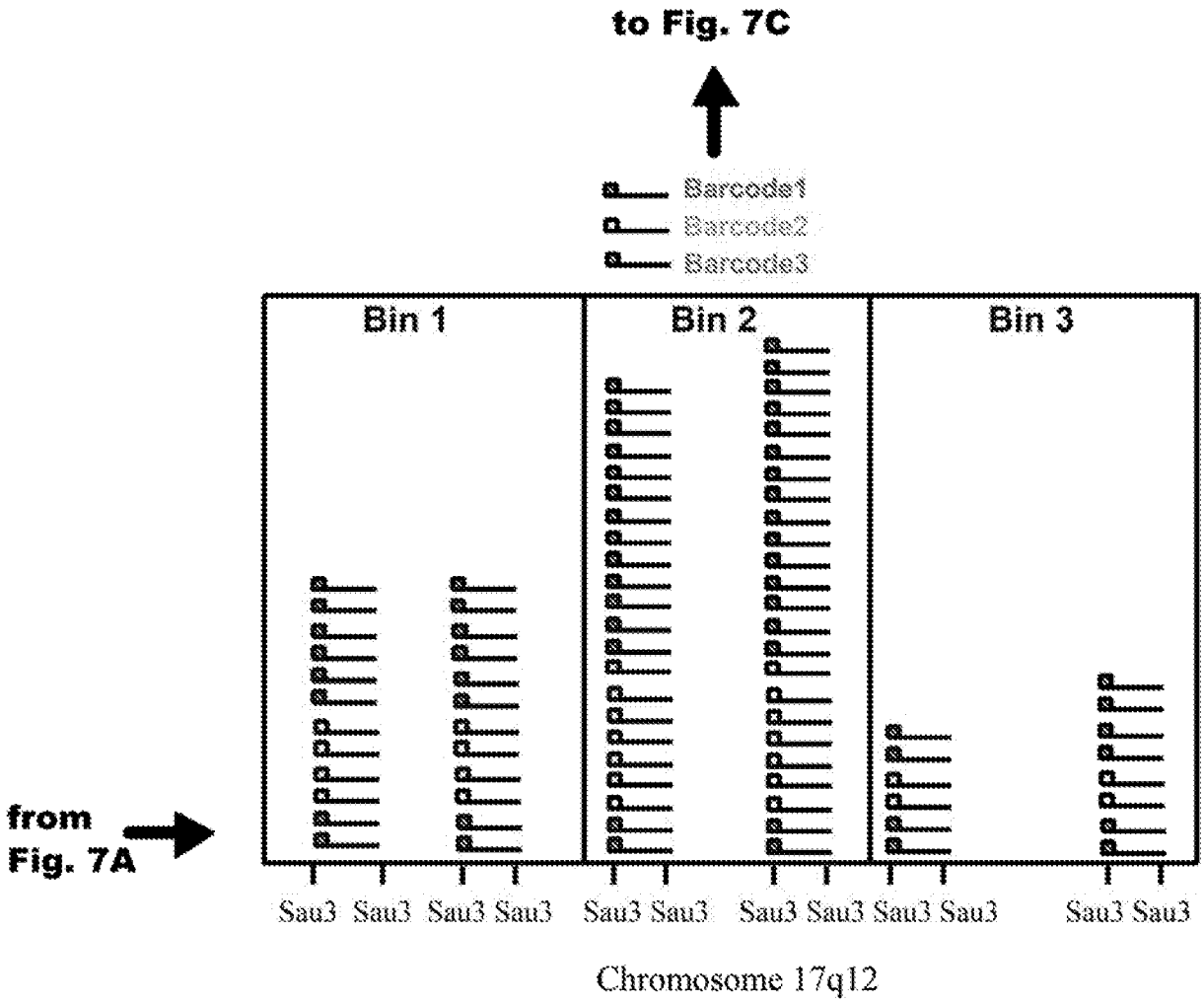
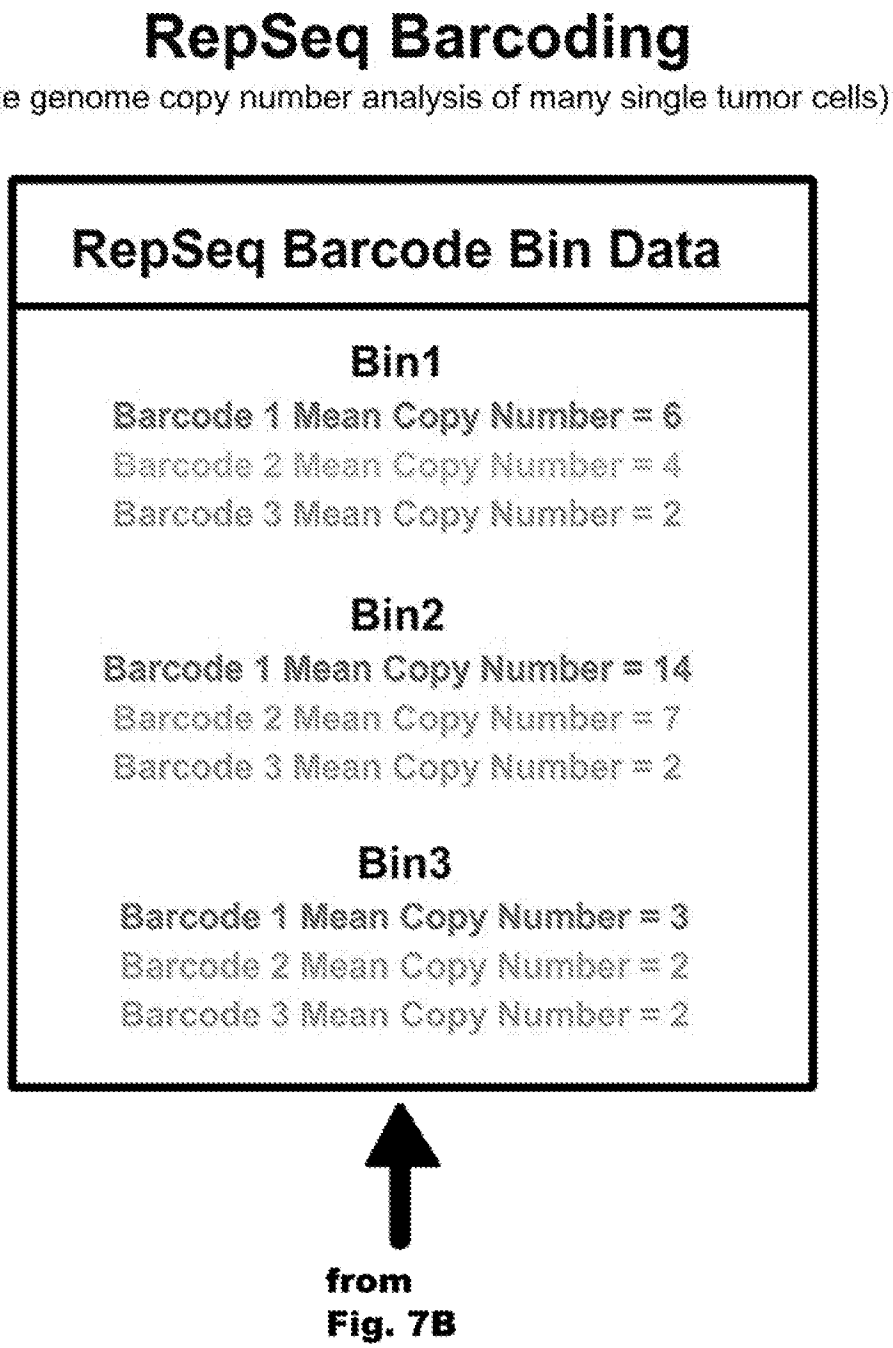


Fig. 7C (continued from Fig. 7B)



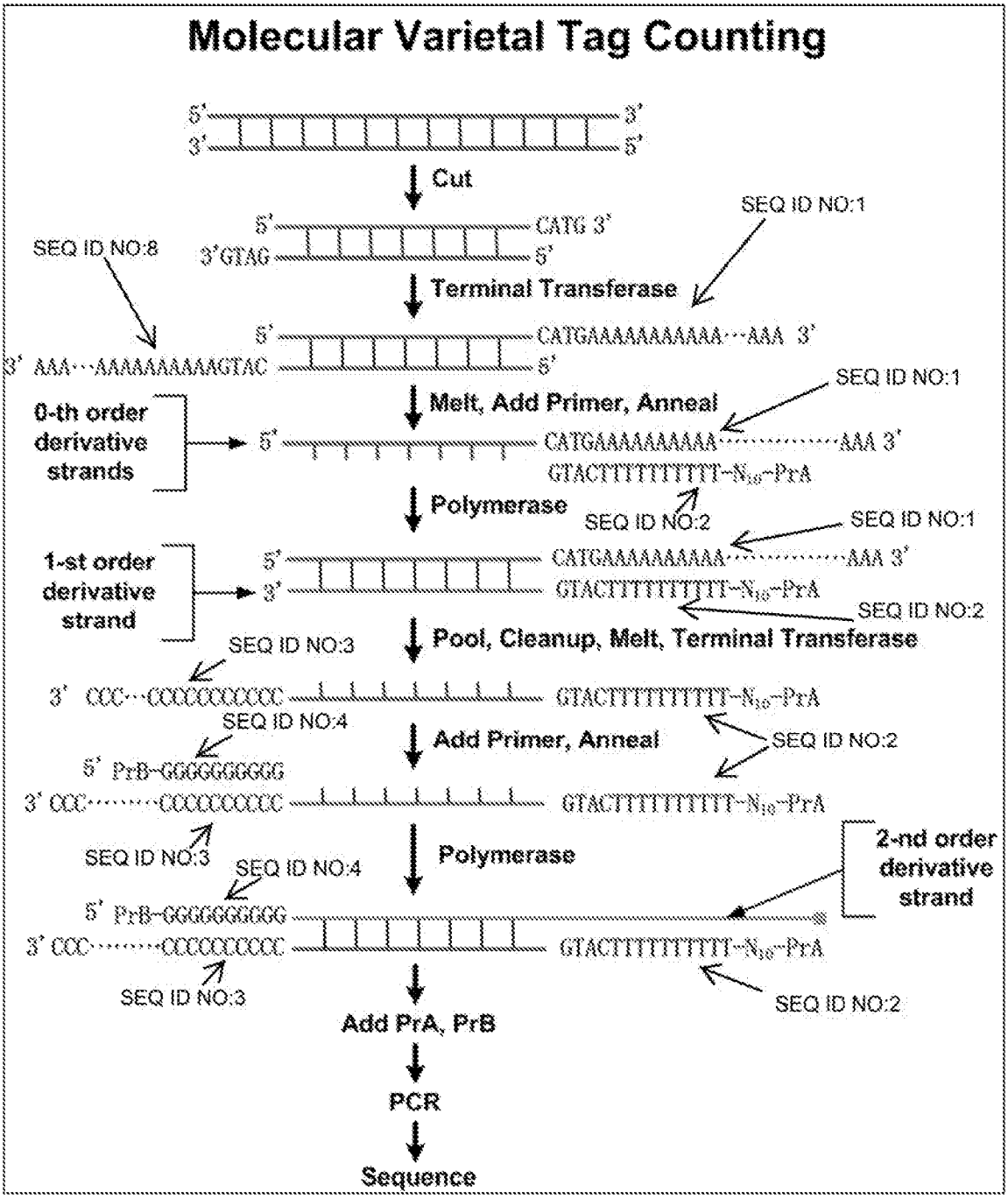
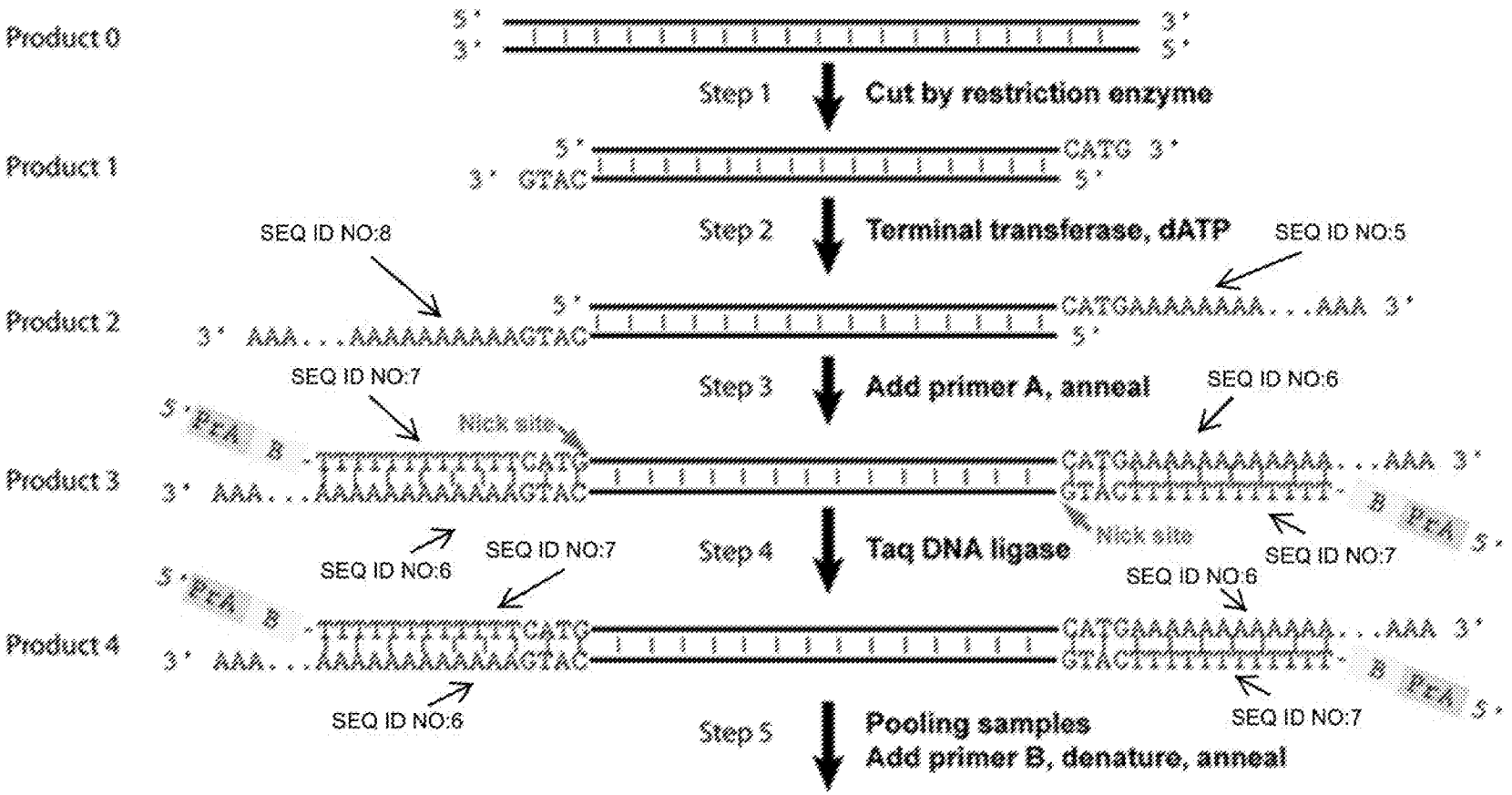

Molecular Varietal Tag Counting
Cut
SEQ ID NO:1
SEQ ID NO:8
Terminal Transferase
Melt, Add Primer, Anneal
0-th order derivative strands
Polymerase
SEQ ID NO:2
1-st order derivative strand
Pool, Cleanup, Melt, Terminal Transferase
SEQ ID NO:3
SEQ ID NO:4
Add Primer, Anneal
2-nd order derivative strand
Add PrA, PrB
PCR
Sequence

Fig. 8

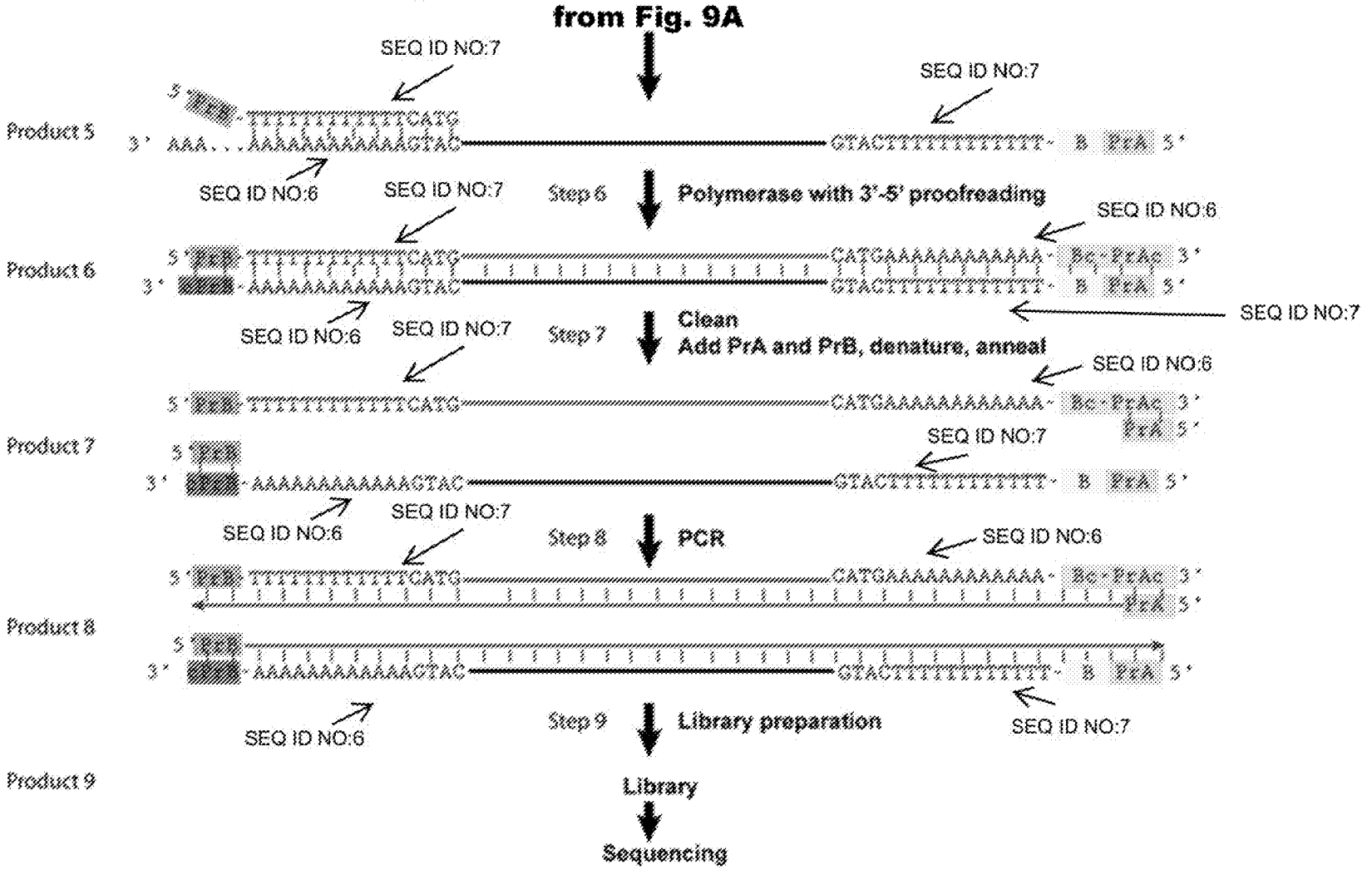
Fig. 9A



to Fig. 9B

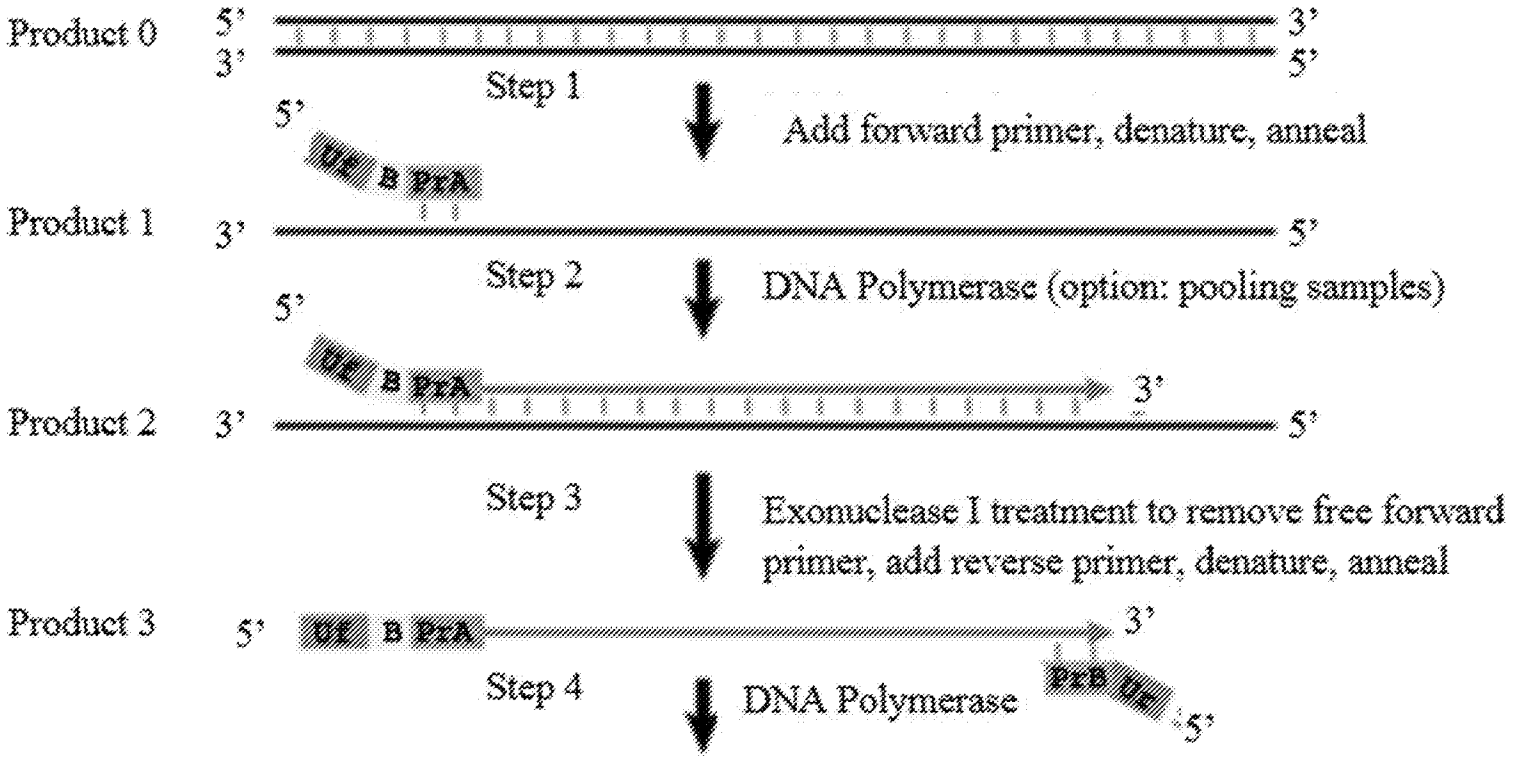
GUARDANT HEALTH EX.1009.426

Fig. 9B (continued from Fig. 9A)



GUARDANT HEALTH EX.1009.427

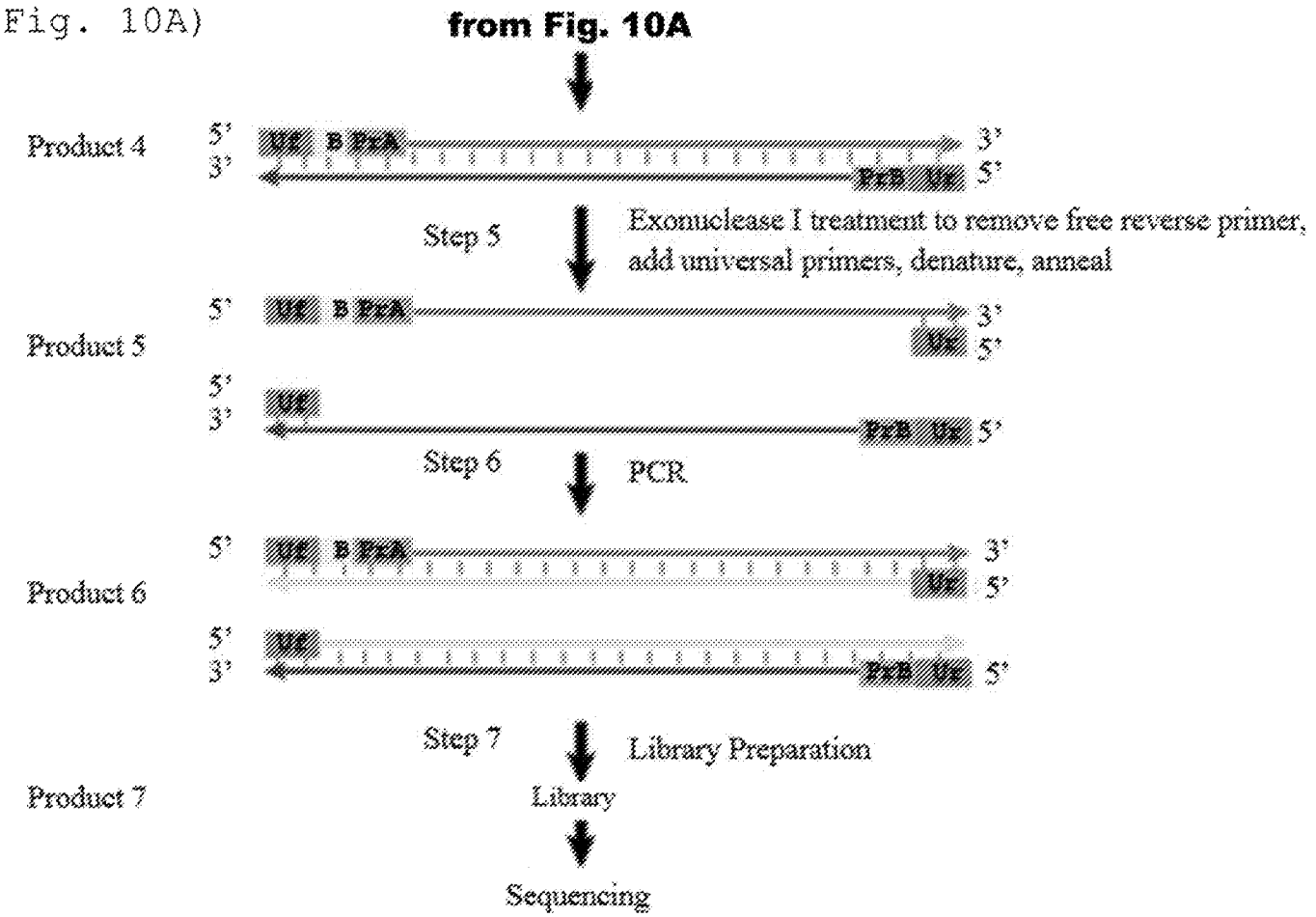
Fig. 10A



to Fig. 10B

Fig. 10B

(continued from Fig. 10A)



This document is scheduled to be published in the Federal Register on 01/07/2019 and available online at https://federalregister.gov/d/2018-28282, and on govinfo.gov

[3510-16-P]

DEPARTMENT OF COMMERCE

United States Patent and Trademark Office

[Docket No. PTO-P-2018-0053]

2019 Revised Patent Subject Matter Eligibility Guidance

AGENCY: United States Patent and Trademark Office, Commerce.

ACTION: Examination Guidance; Request for comments.

SUMMARY: The United States Patent and Trademark Office (USPTO) has prepared revised guidance (2019 Revised Patent Subject Matter Eligibility Guidance) for use by USPTO personnel in evaluating subject matter eligibility. The 2019 Revised Patent Subject Matter Eligibility Guidance revises the procedures for determining whether a patent claim or patent application claim is directed to a judicial exception (laws of nature, natural phenomena, and abstract ideas) under Step 2A of the USPTO's Subject Matter Eligibility Guidance in two ways. First, the 2019 Revised Patent Subject Matter Eligibility Guidance explains that abstract ideas can be grouped as, e.g., mathematical concepts, certain methods of organizing human activity, and mental processes. Second, this guidance explains that a patent claim or patent application claim that recites a judicial exception is not "directed to" the judicial exception if the judicial

GUARDANT HEALTH EX.1009.430

exception is integrated into a practical application of the judicial exception. A claim that recites a judicial exception, but is not integrated into a practical application, is directed to the judicial exception under Step 2A and must then be evaluated under Step 2B (inventive concept) to determine the subject matter eligibility of the claim. The USPTO is seeking public comment on its subject matter eligibility guidance, and particularly the 2019 Revised Patent Subject Matter Eligibility Guidance.

DATES: APPLICABLE DATE: The 2019 Revised Patent Subject Matter Eligibility Guidance is effective on [INSERT DATE OF PUBLICATION IN THE FEDERAL REGISTER]. The 2019 Revised Patent Subject Matter Eligibility Guidance applies to all applications, and to all patents resulting from applications, filed before, on, or after [INSERT DATE OF PUBLICATION IN THE FEDERAL REGISTER].

COMMENT DEADLINE DATE: Written comments must be received on or before [INSERT DATE 60 DAYS AFTER PUBLICATION IN THE FEDERAL REGISTER].

ADDRESSES: Comments must be sent by electronic mail message over the Internet addressed to: Eligibility2019@uspto.gov.

Electronic comments submitted in plain text are preferred, but also may be submitted in ADOBE® portable document format or MICROSOFT WORD® format. Comments not submitted electronically should be submitted on paper in a format that facilitates convenient digital scanning into ADOBE® portable document format. The comments will be available for

GUARDANT HEALTH EX.1009.431

viewing via the USPTO's Internet Web site (http://www.uspto.gov). Because comments will be made available for public inspection, information that the submitter does not desire to make public, such as an address or phone number, should not be included in the comments.

FOR FURTHER INFORMATION CONTACT: June E. Cohan, Senior Legal Advisor, at 571-272-7744 or Carolyn Kosowski, Senior Legal Advisor, at 571–272–7688, both with the Office of Patent Legal Administration.

SUPPLEMENTARY INFORMATION: Patent subject matter eligibility under 35 U.S.C. 101 has been the subject of much attention over the past decade. Recently, much of that attention has focused on how to apply the U.S. Supreme Court's framework for evaluating eligibility (often called the *Alice/Mayo* test).[1] Properly applying the *Alice/Mayo* test in a consistent manner has proven to be difficult, and has caused uncertainty in this area of the law. Among other things, it has become difficult in some cases for inventors, businesses, and other patent stakeholders to reliably and predictably determine what subject matter is patent-eligible. The legal uncertainty surrounding Section 101 poses unique challenges for the USPTO, which must ensure that its more than 8500 patent examiners and administrative patent judges apply the *Alice/Mayo* test in a manner that produces reasonably consistent and predictable results across applications, art units and technology fields.

Since the *Alice/Mayo* test was announced and began to be extensively applied, the courts and the USPTO have tried to consistently distinguish between patent-eligible subject matter and subject

[1] *Alice Corp. Pty. Ltd. v. CLS Bank Int'l*, 573 U.S. 208, 217-18 (2014) (citing *Mayo Collaborative Servs. v. Prometheus Labs., Inc.*, 566 U.S. 66 (2012)).

GUARDANT HEALTH EX.1009.432

matter falling within a judicial exception. Even so, patent stakeholders have expressed a need for more clarity and predictability in its application. In particular, stakeholders have expressed concern with the proper scope and application of the "abstract idea" exception. Some courts share these concerns, for example as demonstrated by several recent concurrences and dissents in the U.S. Court of Appeals for the Federal Circuit ("Federal Circuit") calling for changes in the application of Section 101 jurisprudence.[2] Many stakeholders, judges, inventors, and practitioners across the spectrum have argued that something needs to be done to increase clarity and consistency in how Section 101 is currently applied.

To address these and other concerns, the USPTO is revising its examination procedure with respect to the first step of the *Alice/Mayo* test[3] (Step 2A of the USPTO's Subject Matter Eligibility Guidance as incorporated into the Manual of Patent Examining Procedure ("MPEP") 2106)[4] by: (1) providing groupings of subject matter that is considered an abstract idea; and (2) clarifying that a claim is not "directed to" a judicial exception if the judicial exception is integrated into a practical application of that exception.

Section I of this 2019 Revised Patent Subject Matter Eligibility Guidance explains that the judicial exceptions are for subject matter that has been identified as the "basic tools of scientific

---

[2] *See, e.g., Interval Licensing LLC, v. AOL, Inc.*, 896 F.3d 1335, 1348 (Fed. Cir. 2018) (Plager, J., concurring in part and dissenting in part); *Smart Sys. Innovations, LLC v. Chicago Transit Auth.*, 873 F.3d 1364, 1377 (Fed. Cir. 2017) (Linn, J., dissenting in part and concurring in part); *Berkheimer v. HP Inc.*, 890 F.3d 1369, 1376 (Fed. Cir. 2018) (Lourie, J., joined by Newman, J., concurring in denial of rehearing en banc).

[3] The first step of the *Alice/Mayo* test is to determine whether the claims are "directed to" a judicial exception. *Alice*, 573 U.S. at 217 (citing *Mayo*, 566 U.S. at 77).

[4] All references to the MPEP in the 2019 Revised Patent Subject Matter Eligibility Guidance are to the Ninth Edition, Revision 08-2017 (rev. Jan. 2018), unless otherwise indicated.

GUARDANT HEALTH EX.1009.433

and technological work,"[5] which includes "abstract ideas" such as mathematical concepts, certain methods of organizing human activity, and mental processes; as well as laws of nature and natural phenomena. Only when a claim recites a judicial exception does the claim require further analysis in order to determine its eligibility. The groupings of abstract ideas contained in this guidance enable USPTO personnel to more readily determine whether a claim recites subject matter that is an abstract idea.

Section II explains that the USPTO has set forth a revised procedure, rooted in Supreme Court caselaw, to determine whether a claim is "directed to" a judicial exception under the first step of the *Alice/Mayo* test (USPTO Step 2A).

Section III explains the revised procedure that will be applied by the USPTO. The procedure focuses on two aspects of Revised Step 2A: (1) whether the claim recites a judicial exception; and (2) whether a recited judicial exception is integrated into a practical application. Only when a claim recites a judicial exception and fails to integrate the exception into a practical application, is the claim "directed to" a judicial exception, thereby triggering the need for further analysis pursuant to the second step of the *Alice/Mayo* test (USPTO Step 2B). Finally, if further analysis at Step 2B is needed (for example to determine whether the claim merely recites well-understood, routine, conventional activity), this 2019 Revised Patent Subject Matter Eligibility

[5] *Mayo*, 566 U.S. at 71 ("Phenomena of nature, though just discovered, mental processes, and abstract intellectual concepts are not patentable, as they are the basic tools of scientific and technological work" (quoting *Gottschalk v. Benson*, 409 U.S. 63, 67 (1972)).

GUARDANT HEALTH EX.1009.434

Guidance explains that the examiner or administrative patent judge will proceed in accordance with existing USPTO guidance as modified in April 2018.[6]

The USPTO is seeking public comment on its subject matter eligibility guidance, and particularly the 2019 Revised Patent Subject Matter Eligibility Guidance. The USPTO is determined to continue its mission to provide predictable and reliable patent rights in accordance with this rapidly evolving area of the law. The USPTO's ultimate goal is to draw distinctions between claims to principles in the abstract and claims that integrate those principles into a practical application. To that end, the USPTO may issue further guidance, or modify the current guidance, in the future based on its review of the comments received, further experience of the USPTO and its stakeholders, and additional judicial actions. Implementation of examination guidance on eligibility is an iterative process and may continue with periodic supplements. The USPTO invites the public to submit suggestions on eligibility-related topics to address in future guidance supplements as part of their comments on the USPTO's subject matter eligibility guidance.

***Impact on Examination Procedure and Prior Examination Guidance*:** This 2019 Revised Patent Subject Matter Eligibility Guidance supersedes MPEP 2106.04(II) (Eligibility Step 2A: Whether a Claim Is Directed to a Judicial Exception) to the extent it equates claims "reciting" a judicial exception with claims "directed to" a judicial exception, along with any other portion of the MPEP that conflicts with this guidance. A chart identifying portions of the MPEP that are

[6] USPTO Memorandum of April 19, 2018, "Changes in Examination Procedure Pertaining to Subject Matter Eligibility, Recent Subject Matter Eligibility Decision (*Berkheimer v. HP, Inc.*)" (Apr. 19, 2018), *available at* https://www.uspto.gov/sites/default/files/documents/memo-berkheimer-20180419.PDF [hereinafter "USPTO *Berkheimer* Memorandum"].

GUARDANT HEALTH EX.1009.435

affected by this guidance will be available for viewing via the USPTO's Internet Web site (http://www.uspto.gov). This 2019 Revised Patent Subject Matter Eligibility Guidance also supersedes all versions of the USPTO's "Eligibility Quick Reference Sheet Identifying Abstract Ideas" (first issued in July 2015 and updated most recently in July 2018). Eligibility-related guidance issued prior to the Ninth Edition, R-08.2017, of the MPEP (published Jan. 2018) should not be relied upon. However, any claim considered patent eligible under prior guidance should be considered patent eligible under this guidance.

This guidance does not constitute substantive rulemaking and does not have the force and effect of law. The guidance sets out agency policy with respect to the USPTO's interpretation of the subject matter eligibility requirements of 35 U.S.C. 101 in view of decisions by the Supreme Court and the Federal Circuit. The guidance was developed as a tool for internal USPTO management and does not create any right or benefit, substantive or procedural, enforceable by any party against the USPTO. Rejections will continue to be based upon the substantive law, and it is those rejections that are appealable to the Patent Trial and Appeal Board (PTAB) and the courts. All USPTO personnel are, as a matter of internal agency management, expected to follow the guidance. Failure of USPTO personnel to follow the guidance, however, is not, in itself, a proper basis for either an appeal or a petition.

I. GROUPINGS OF ABSTRACT IDEAS

The Supreme Court has held that the patent eligibility statute, Section 101, contains an implicit exception for "[l]aws of nature, natural phenomena, and abstract ideas," which are "the basic

GUARDANT HEALTH EX.1009.436

tools of scientific and technological work."[7] Yet, the Court has explained that "[a]t some level, all inventions embody, use, reflect, rest upon, or apply laws of nature, natural phenomena, or abstract ideas," and has cautioned "to tread carefully in construing this exclusionary principle lest it swallow all of patent law."[8]

Since the *Alice* case, courts have been "compare[ing] claims at issue to those claims already found to be directed to an abstract idea in previous cases."[9] Likewise, the USPTO has issued guidance to the patent examining corps about Federal Circuit decisions applying the *Alice/Mayo* test, for instance describing the subject matter claimed in the patent in suit and noting whether or not certain subject matter has been identified as an abstract idea.[10]

While that approach was effective soon after *Alice* was decided, it has since become impractical. The Federal Circuit has now issued numerous decisions identifying subject matter as abstract or

---

[7] *Alice Corp.*, 573 U.S. at 216 (internal citation and quotation marks omitted); *Mayo*, 566 U.S. at 71.

[8] *Id.* (internal citation and quotation marks omitted).

[9] *See Enfish, LLC v. Microsoft Corp.*, 822 F.3d 1327, 1334 (Fed. Cir. 2016); *see also Amdocs (Israel) Ltd. v. Openet Telecom, Inc.*, 841 F.3d 1288, 1294 (Fed. Cir. 2016) ("[T]he decisional mechanism courts now apply [to identify an abstract idea] is to examine earlier cases in which a similar or parallel descriptive nature can be seen—what prior cases were about, and which way they were decided.").

[10] *See, e.g.*, 2014 Interim Guidance on Subject Matter Eligibility, 79 Fed. Reg. 74618, 74628-32 (Dec. 16, 2014) (discussing concepts identified as abstract ideas); July 2015 Update: Subject Matter Eligibility (Jul. 30, 2015), at 3-5, *available at* https://www.uspto.gov/sites/default/files/documents/ieg-july-2015-update.pdf (same); USPTO Memorandum of May 19, 2016, "Recent Subject Matter Eligibility Decisions (*Enfish, LLC v. Microsoft Corp.* and *TLI Communications LLC v. A.V. Automotive, LLC*)," at 2 (May 19, 2016), *available at* https://www.uspto.gov/sites/default/files/documents/ieg-may-2016_enfish_memo.pdf [hereinafter, "USPTO *Enfish* Memorandum"] (discussing the abstract idea in *TLI Communications LLC v. A.V. Automotive, LLC*, 823 F.3d 607 (Fed. Cir. 2016)); USPTO Memorandum of November 2, 2016, "Recent Subject Matter Eligibility Decisions," at 2 (Nov. 2, 2016), *available at* https://www.uspto.gov/sites/default/files/documents/McRo-Bascom-Memo.pdf [hereinafter, "USPTO *McRo* Memorandum"] (discussing how the claims in *McRO, Inc. v. Bandai Namco Games America Inc.*, 837 F.3d 1299 (Fed. Cir. 2016), were directed to an improvement instead of an abstract idea); USPTO Memorandum of April 2, 2018, "Recent Subject Matter Eligibility Decisions" (Apr. 2, 2018), *available at* https://www.uspto.gov/sites/default/files/documents/memo-recent-sme-ctdec-20180402.PDF [hereinafter "USPTO *Finjan* Memorandum"] (discussing how the claims in *Finjan Inc. v. Blue Coat Systems, Inc.*, 879 F.3d 1299 (Fed. Cir. 2018), and *Core Wireless Licensing, S.A.R.L. v. LG Electronics, Inc.*, 880 F.3d 1356 (Fed. Cir. 2018), were directed to improvements instead of abstract ideas); USPTO *Berkheimer* Memorandum at 2 (discussing the abstract idea in *Berkheimer*); MPEP 2106.04(a) (reviewing cases that did and did not identify abstract ideas).

GUARDANT HEALTH EX.1009.437

non-abstract in the context of specific cases, and that number is continuously growing. In addition, similar subject matter has been described both as abstract and not abstract in different cases.[11] The growing body of precedent has become increasingly more difficult for examiners to apply in a predictable manner, and concerns have been raised that different examiners within and between technology centers may reach inconsistent results.

The USPTO, therefore, aims to clarify the analysis. In accordance with judicial precedent and in an effort to improve consistency and predictability, the 2019 Revised Patent Subject Matter Eligibility Guidance extracts and synthesizes key concepts identified by the courts as abstract ideas to explain that the abstract idea exception includes the following groupings of subject matter, when recited as such in a claim limitation(s) (that is, when recited on their own or per se):

a) **Mathematical concepts** – mathematical relationships, mathematical formulas or equations, mathematical calculations;[12]

---

[11] *E.g.*, compare *TLI Commc'ns*, 823 F.3d at 611, with *Enfish*, 822 F.3d at 1335, and *Visual Memory LLC v. NVIDIA Corp.*, 867 F.3d 1253, 1258 (Fed. Cir. 2017). While computer operations such as "output of data analysis . . . can be abstract," *Credit Acceptance Corp. v. Westlake Servs.*, 859 F.3d 1044, 1056 (Fed. Cir. 2017), "software-based innovations can [also] make 'non-abstract improvements to computer technology' and be deemed patent-eligible subject matter at step 1 [of the *Mayo/Alice* test]," *Finjan*, 879 F.3d at 1304 (*quoting Enfish*, 822 F.3d at 1335). Indeed, the Federal Circuit has held that "improvements in computer-related technology" and "claims directed to software" are not "inherently abstract." *Enfish*, 822 F.3d at 1335; *see also Visual Memory*, 867 F.3d at 1258. These developments in the caselaw can create complications for the patent-examination process. For example, claims in one application could be deemed to be abstract, whereas slightly different claims directed to the same or similar subject matter could be determined to reflect a patent eligible "improvement." Alternatively, claims in one application could be found to be abstract, whereas claims to the same or similar subject matter in another application, containing additional or different embodiments in the specification, could be deemed eligible as not directed to an abstract idea. In other words, the finding that the subject matter claimed in a prior patent was "abstract" as claimed may not determine whether similar subject matter in another application, claimed somewhat differently or supported by a different disclosure, is directed to an abstract idea and therefore patent ineligible.

[12] *Bilski v. Kappos*, 561 U.S. 593, 611 (2010) ("The concept of hedging . . . reduced to a mathematical formula . . . is an unpatentable abstract idea[.]"); *Diamond v. Diehr*, 450 U.S. 175, 191 (1981) ("A mathematical formula as such is not accorded the protection of our patent laws") (citing *Benson*, 409 U.S. 63); *Parker v. Flook*, 437 U.S. 584, 594 (1978) ("[T]he discovery of [a mathematical formula] cannot support a patent unless there is some other inventive concept in its application."); *Benson*, 409 U.S. at 71-72 (concluding that permitting a patent on the claimed

GUARDANT HEALTH EX.1009.438

b) **Certain methods of organizing human activity** – fundamental economic principles or practices (including hedging, insurance, mitigating risk); commercial or legal interactions (including agreements in the form of contracts; legal obligations; advertising, marketing or sales activities or behaviors; business relations); managing personal behavior or relationships or interactions between people (including social activities, teaching, and following rules or instructions);[13] and

---

invention "would wholly pre-empt the mathematical formula and in practical effect would be a patent on the algorithm itself"); *Mackay Radio & Telegraph Co. v. Radio Corp. of Am.*, 306 U.S. 86, 94 (1939) ("[A] scientific truth, or the mathematical expression of it, is not patentable invention[.]"); *SAP America, Inc. v. InvestPic, LLC*, 898 F.3d 1161, 1163 (Fed. Cir. 2018) (holding that claims to a "series of mathematical calculations based on selected information" are directed to abstract ideas); *Digitech Image Techs., LLC v. Elecs. for Imaging, Inc.*, 758 F.3d 1344, 1350 (Fed. Cir. 2014) (holding that claims to a "process of organizing information through mathematical correlations" are directed to an abstract idea); *Bancorp Servs., LLC v. Sun Life Assurance Co. of Can. (U.S.)*, 687 F.3d 1266, 1280 (Fed. Cir. 2012) (identifying the concept of "managing a stable value protected life insurance policy by performing calculations and manipulating the results" as an abstract idea).

[13] *Alice*, 573 U.S. at 219-20 (concluding that use of a third party to mediate settlement risk is a "fundamental economic practice" and thus an abstract idea); *id.* (describing the concept of risk hedging identified as an abstract idea in *Bilski* as "a method of organizing human activity"); *Bilski*, 561 U.S. at 611-612 (concluding that hedging is a "fundamental economic practice" and therefore an abstract idea); *Bancorp*, 687 F.3d at 1280 (concluding that "managing a stable value protected life insurance policy by performing calculations and manipulating the results" is an abstract idea); *Inventor Holdings, LLC v. Bed Bath & Beyond, Inc.*, 876 F.3d 1372, 1378-79 (Fed. Cir. 2017) (holding that concept of "local processing of payments for remotely purchased goods" is a "fundamental economic practice, which *Alice* made clear is, without more, outside the patent system."); *OIP Techs., Inc. v. Amazon.com, Inc.*, 788 F.3d 1359, 1362-63 (Fed. Cir. 2015) (concluding that claimed concept of "offer-based price optimization" is an abstract idea "similar to other 'fundamental economic concepts' found to be abstract ideas by the Supreme Court and this court"); *buySAFE, Inc. v. Google, Inc.*, 765 F.3d. 1350, 1355 (Fed. Cir. 2014) (holding that concept of "creating a contractual relationship—a 'transaction performance guaranty'" is an abstract idea); *In re Comiskey*, 554 F.3d 967, 981 (Fed Cir. 2009) (claims directed to "resolving a legal dispute between two parties by the decision of a human arbitrator" are ineligible); *Ultramercial, Inc. v. Hulu*, LLC, 772 F.3d 709, 715 (Fed Cir. 2014) (holding that claim "describe[ing] only the abstract idea of showing an advertisement before delivering free content" is patent ineligible); *In re Ferguson*, 558 F.3d 1359, 1364 (Fed Cir. 2009) (holding methods "directed to organizing business or legal relationships in the structuring of a sales force (or marketing company)" to be ineligible); *Credit Acceptance*, 859 F.3d 1044 at 1054 ("The Board determined that the claims are directed to the abstract idea of 'processing an application for financing a purchase.' . . . We agree."); *Interval Licensing*, 896 F.3d at 1344–45 (concluding that "[s]tanding alone, the act of providing someone an additional set of information without disrupting the ongoing provision of an initial set of information is an abstract idea," observing that the district court "pointed to the nontechnical human activity of passing a note to a person who is in the middle of a meeting or conversation as further illustrating the basic, longstanding practice that is the focus of the [patent ineligible] claimed invention."); *Voter Verified, Inc. v. Election Systems & Software, LLC*, 887 F.3d 1376, 1385 (Fed Cir. 2018) (finding the concept of "voting, verifying the vote, and submitting the vote for tabulation," a "fundamental activity" that humans have performed for hundreds of years, to be an abstract idea); *In re Smith*, 815 F.3d 816, 818 (Fed Cir. 2016) (concluding that "[a]pplicants' claims, directed to rules for conducting a wagering game" are abstract).

GUARDANT HEALTH EX.1009.439

c) **Mental processes** – concepts performed in the human mind[14] (including an observation, evaluation, judgment, opinion).[15]

Claims that do not recite matter that falls within these enumerated groupings of abstract ideas should not be treated as reciting abstract ideas, except as follows: In the rare circumstance in which a USPTO employee believes a claim limitation that does not fall within the enumerated groupings of abstract ideas should nonetheless be treated as reciting an abstract idea, the procedure described in Section III.C for analyzing the claim should be followed.

---

[14] If a claim, under its broadest reasonable interpretation, covers performance in the mind but for the recitation of generic computer components, then it is still in the mental processes category unless the claim cannot practically be performed in the mind. *See Intellectual Ventures I LLC v. Symantec Corp.*, 838 F.3d 1307, 1318 (Fed. Cir. 2016) ("[W]ith the exception of generic computer-implemented steps, there is nothing in the claims themselves that foreclose them from being performed by a human, mentally or with pen and paper."); *Mortg. Grader, Inc. v. First Choice Loan Servs. Inc.*, 811 F.3d. 1314, 1324 (Fed. Cir. 2016) (holding that computer-implemented method for "anonymous loan shopping" was an abstract idea because it could be "performed by humans without a computer"); *Versata Dev. Grp. v. SAP Am., Inc.*, 793 F.3d 1306, 1335 (Fed Cir. 2015) ("Courts have examined claims that required the use of a computer and still found that the underlying, patent-ineligible invention could be performed via pen and paper or in a person's mind."); *CyberSource Corp. v. Retail Decisions, Inc.*, 654 F.3d 1366, 1375, 1372 (Fed Cir. 2011) (holding that the incidental use of "computer" or "computer readable medium" does not make a claim otherwise directed to process that "can be performed in the human mind, or by a human using a pen and paper" patent eligible); *id.* at 1376 (distinguishing *Research Corp. Techs. v. Microsoft Corp.*, 627 F.3d 859 (Fed. Cir. 2010), and *SiRF Tech., Inc. v. Int'l Trade Comm'n*, 601 F.3d 1319 (Fed. Cir. 2010), as directed to inventions that "could not, as a practical matter, be performed entirely in a human's mind"). Likewise, performance of a claim limitation using generic computer components does not necessarily preclude the claim limitation from being in the mathematical concepts grouping, *Benson*, 409 U.S. at 67, or the certain methods of organizing human activity grouping, *Alice*, 573 U.S. at 219-20.

[15] *Mayo*, 566 U.S. at 71 ("'[M]ental processes[] and abstract intellectual concepts are not patentable, as they are the basic tools of scientific and technological work'" (quoting *Benson*, 409 U.S. at 67)); *Flook*, 437 U.S. at 589 (same); *Benson*, 409 U.S. at 67, 65 (noting that the claimed "conversion of [binary-coded decimal] numerals to pure binary numerals can be done mentally," i.e., "as a person would do it by head and hand."); *Synopsys, Inc. v. Mentor Graphics Corp.*, 839 F.3d 1138, 1139, (Fed. Cir. 2016) (holding that claims to the mental process of "translating a functional description of a logic circuit into a hardware component description of the logic circuit" are directed to an abstract idea, because the claims "read on an individual performing the claimed steps mentally or with pencil and paper"); *Mortg. Grader*, 811 F.3d. at 1324 (concluding that concept of "anonymous loan shopping" is an abstract idea because it could be "performed by humans without a computer"); *In re BRCA1 & BRCA2-Based Hereditary Cancer Test Patent Litig.*, 774 F.3d 755, 763 (Fed. Cir. 2014) (concluding that concept of "comparing BRCA sequences and determining the existence of alterations" is an "abstract mental process"); *In re Brown*, 645 F. App'x. 1014, 1017 (Fed Cir. 2016) (non-precedential) (claim limitations "encompass the mere idea of applying different known hair styles to balance one's head. Identifying head shape and applying hair designs accordingly is an abstract idea capable, as the Board notes, of being performed entirely in one's mind").

GUARDANT HEALTH EX.1009.440

II. "DIRECTED TO" A JUDICIAL EXCEPTION

The Supreme Court has long distinguished between principles themselves (which are not patent eligible) and the integration of those principles into practical applications (which are patent eligible).[16] Similarly, in a growing body of decisions, the Federal Circuit has distinguished between claims that are "directed to" a judicial exception (which require further analysis to determine their eligibility) and those that are not (which are therefore patent eligible). [17] For example, an improvement in the functioning of a computer or other technology or technological field may render a claim patent eligible at step one of the *Alice/Mayo* test even if it recites an abstract idea, law of nature, or natural phenomenon.[18] Moreover, recent Federal Circuit jurisprudence has indicated that eligible subject matter can often be identified either at the first or

---

[16] *See, e.g., Alice*, 573 U.S. at 217 (explaining that "in applying the §101 exception, we must distinguish between patents that claim the 'buildin[g] block[s]' of human ingenuity and those that integrate the building blocks into something more" (quoting *Mayo*, 566 U.S. at 89) and stating that *Mayo* "set forth a framework for distinguishing patents that claim laws of nature, natural phenomena, and abstract ideas from those that claim patent-eligible applications of those concepts"); *Mayo*, 566 U.S. at 80, 84 (noting that the Court in *Diehr* found "the overall process patent eligible because of the way the additional steps of the process integrated the equation into the process as a whole," but the Court in *Benson* "held that simply implementing a mathematical principle on a physical machine, namely a computer, was not a patentable application of that principle"); *Bilski*, 561 U.S. at 611 ("*Diehr* explained that while an abstract idea, law of nature, or mathematical formula could not be patented, 'an *application* of a law of nature or mathematical formula to a known structure or process may well be deserving of patent protection.'" (quoting *Diehr*, 450 U.S. at 187) (emphasis in original)); *Diehr*, 450 U.S. at 187, 192 n.14 (explaining that the process in *Flook* was ineligible not because it contained a mathematical formula, but because it did not provide an application of the formula); *Mackay Radio*, 306 U.S. at 94 ("While a scientific truth, or the mathematical expression of it, is not patentable invention, a novel and useful structure created with the aid of knowledge of scientific truth may be."); *Le Roy v. Tatham*, 55 U.S. (14 How.) 156, 175 (1852) ("The elements of the [natural phenomena] exist; the invention is not in discovering them, but in applying them to useful objects.").

[17] *See, e.g.*, MPEP 2106.06(b) (summarizing *Enfish*, *McRO*, and other cases that were eligible as improvements to technology or computer functionality instead of abstract ideas); USPTO *Finjan* Memorandum (discussing *Finjan*, and *Core Wireless*); USPTO Memorandum of June 7, 2018, "Recent Subject Matter Eligibility Decision: *Vanda Pharmaceuticals Inc. v. West-Ward Pharmaceuticals*," *available at* https://www.uspto.gov/sites/default/files/documents/memo-vanda-20180607.PDF [hereinafter "USPTO *Vanda* Memorandum"]; *BASCOM Glob. Internet Servs., Inc. v. AT&T Mobility LLC*, 827 F.3d 1341, 1352 (Fed. Cir. 2016) (concluding that claims could be eligible if ordered combination of limitations "transform the abstract idea … into a particular, practical application of that abstract idea."); *Arrhythmia Research Tech., Inc. v. Corazonix Corp.*, 958 F.2d 1053, 1056–57 (Fed. Cir. 1992) ("As the jurisprudence developed, inventions that were implemented by the mathematically-directed performance of computers were viewed in the context of the practical application to which the computer-generated data were put."); *CLS Bank Int'l v. Alice Corp. Pty. Ltd.*, 717 F.3d 1269, 1315 (Fed. Cir. 2013) (Moore, J., joined by Rader, C.J., and Linn and O'Malley, JJ., dissenting in part) ("The key question is thus whether a claim recites a sufficiently concrete and practical application of an abstract idea to qualify as patent-eligible."), *aff'd*, 573 U.S. 208(2014).

[18] *See, e.g., McRO*, 837 F.3d at 1316; *Enfish*, 822 F.3d at 1336; *Core Wireless*, 880 F.3d at 1362.

GUARDANT HEALTH EX.1009.441

the second step of the *Alice/Mayo* test.[19] These revised patent examination procedures are designed to more accurately and consistently identify claims that recite a practical application of a judicial exception (and thus are not "directed to" a judicial exception), thereby increasing predictability and consistency in the patent eligibility analysis. This analysis is performed at USPTO Step 2A, and incorporates certain considerations that have been applied by the courts at step one and at step two of the *Alice/Mayo* framework, given the recognized overlap in the steps depending on the facts of any given case.

In accordance with judicial precedent, and to increase consistency in examination practice, the 2019 Revised Patent Subject Matter Eligibility Guidance sets forth a procedure to determine whether a claim is "directed to" a judicial exception under USPTO Step 2A. Under the procedure, if a claim recites a judicial exception (a law of nature, a natural phenomenon, or an abstract idea as grouped in Section I, above), it must then be analyzed to determine whether the recited judicial exception is integrated into a practical application of that exception. A claim is not "directed to" a judicial exception, and thus is patent eligible, if the claim as a whole integrates the recited judicial exception into a practical application of that exception. A claim

---

[19] *See, e.g., Vanda Pharm. Inc. v. West-Ward Pharm. Int'l Ltd.*, 887 F.3d 1117, 1134 (Fed. Cir. 2018) ("If the claims are not directed to a patent ineligible concept at step one, we need not address step two of the inquiry."); *Rapid Litig. Mgmt. Ltd. v. CellzDirect, Inc.*, 827 F.3d 1042, 1050 (Fed. Cir. 2016) (holding that claimed invention is patent eligible because it is not directed to a patent-ineligible concept under step one or is an inventive application of the patent-ineligible concept under step two); *Enfish*, 822 F.3d at 1339 (noting that eligibility determination can be reached either because claims not directed to an abstract idea under step one or recite a concrete improvement under step two); *McRO*, 837 F.3d at 1313 (recognizing that the "court must look to the claims as an ordered combination" in determining patentability "[w]hether at step one or step two of the *Alice* test"); *Amdocs*, 841 F.3d at 1294 (observing that recent cases "suggest that there is considerable overlap between step one and step two, and in some situations [the inventive concept] analysis could be accomplished without going beyond step one"). *See also Ancora Techs. v. HTC Am.*, 908 F.3d 1343, 1349 (Fed. Cir. 2018) (noting, in accord with the "recognition of overlaps between some step one and step two considerations," that its conclusion of eligibility at step one is "indirectly reinforced by some of [its] prior holdings under step two").

GUARDANT HEALTH EX.1009.442

that integrates a judicial exception into a practical application will apply, rely on, or use the judicial exception in a manner that imposes a meaningful limit on the judicial exception, such that the claim is more than a drafting effort designed to monopolize the judicial exception.

III. INSTRUCTIONS FOR APPLYING REVISED STEP 2A DURING EXAMINATION

Examiners should determine whether a claim satisfies the criteria for subject matter eligibility by evaluating the claim in accordance with the criteria discussed in MPEP 2106, i.e., whether the claim is to a statutory category (Step 1) and the *Alice/Mayo* test for judicial exceptions (Steps 2A and 2B). The procedure set forth herein (referred to as "revised Step 2A") changes how examiners should apply the first step of the *Alice/Mayo* test, which determines whether a claim is "directed to" a judicial exception.

As before, Step 1 of the USPTO's eligibility analysis entails considering whether the claimed subject matter falls within the four statutory categories of patentable subject matter identified by 35 U.S.C. 101: process, machine, manufacture, or composition of matter. The 2019 Revised Patent Subject Matter Eligibility Guidance does not change Step 1 or the streamlined analysis, which are discussed in MPEP 2106.03 and 2106.06, respectively. Examiners may continue to use a streamlined analysis (Pathway A) when the patent eligibility of a claim is self-evident.

GUARDANT HEALTH EX.1009.443

Step 2A of the 2019 Revised Patent Subject Matter Eligibility Guidance is a two-prong inquiry. In Prong One, examiners evaluate whether the claim recites a judicial exception.[20] This prong is similar to procedures in prior guidance except that when determining if a claim recites an abstract idea, examiners now refer to the subject matter groupings of abstract ideas in Section I instead of comparing the claimed concept to the USPTO's prior "Eligibility Quick Reference Sheet Identifying Abstract Ideas."

- If the claim recites a judicial exception (i.e., an abstract idea enumerated in Section I of the 2019 Revised Patent Subject Matter Eligibility Guidance, a law of nature, or a natural phenomenon), the claim requires further analysis in Prong Two.
- If the claim does not recite a judicial exception (a law of nature, natural phenomenon, or subject matter within the enumerated groupings of abstract ideas in Section I), then the claim is eligible at Prong One of revised Step 2A. This concludes the eligibility analysis, except in the rare circumstance described below.[21]
- In the rare circumstance in which an examiner believes a claim limitation that does not fall within the enumerated groupings of abstract ideas should nonetheless be treated as reciting an abstract idea, the procedure described in Section III.C for analyzing the claim should be followed.

---

[20] This notice does not change the type of claim limitations that are considered to recite a law of nature or natural phenomenon. For more information about laws of nature and natural phenomena, including products of nature, see MPEP 2106.04(b) and (c).

[21] Even if a claim is determined to be patent eligible under section 101, this or any other step of the eligibility analysis does not end the inquiry. The claims must also satisfy the other conditions and requirements for patentability, for example, under section 102 (novelty), 103 (nonobviousness), or 112 (enablement, written description, definiteness). *Bilski*, 561 U.S. at 602. Examiners should take care not to confuse or intermingle patentability requirements of these separate sections with patent eligibility analysis under section 101.

GUARDANT HEALTH EX.1009.444

In Prong Two, examiners evaluate whether the claim recites additional elements that integrate the exception into a practical application of that exception. This prong adds a more detailed eligibility analysis to step one of the *Alice/Mayo* test (USPTO Step 2A) than was required under prior guidance.

- If the recited exception is integrated into a practical application of the exception, then the claim is eligible at Prong Two of revised Step 2A. This concludes the eligibility analysis.
- If, however, the additional elements do not integrate the exception into a practical application, then the claim is directed to the recited judicial exception, and requires further analysis under Step 2B (where it may still be eligible if it amounts to an "inventive concept").[22]

The following discussion provides additional detail on this revised procedure.

A. Revised Step 2A

1. Prong One: Evaluate Whether The Claim Recites A Judicial Exception

In Prong One, examiners should evaluate whether the claim recites a judicial exception, i.e., an abstract idea, a law of nature, or a natural phenomenon. If the claim does not recite a judicial exception, it is not directed to a judicial exception (Step 2A: NO) and is eligible. This concludes the eligibility analysis. If the claim does recite a judicial exception, then it requires further analysis in Prong Two of Revised Step 2A to determine whether it is directed to the recited

[22] *See, e.g.*, *Amdocs*, 841 F.3d at 1300, 1303; *BASCOM*, 827 F.3d at 1349-52; *DDR Holdings, LLC v. Hotels.com*, L.P., 773 F.3d 1245, 1257-59 (Fed. Cir. 2014); USPTO *Berkheimer* Memorandum; *see also Rapid Litig.*, 827 F.3d at 1050 (holding that claimed invention is patent eligible because it is not directed to a patent-ineligible concept under step one or is an inventive application of the patent-ineligible concept under step two).

GUARDANT HEALTH EX.1009.445

exception, as explained in Section III.A.2 of the 2019 Revised Patent Subject Matter Eligibility Guidance.

For abstract ideas, Prong One represents a change as compared to prior guidance. To determine whether a claim recites an abstract idea in Prong One, examiners are now to: (a) identify the specific limitation(s) in the claim under examination (individually or in combination) that the examiner believes recites an abstract idea; and (b) determine whether the identified limitation(s) falls within the subject matter groupings of abstract ideas enumerated in Section I of the 2019 Revised Patent Subject Matter Eligibility Guidance. If the identified limitation(s) falls within the subject matter groupings of abstract ideas enumerated in Section I, analysis should proceed to Prong Two in order to evaluate whether the claim integrates the abstract idea into a practical application. When evaluating Prong One, examiners are no longer to use the USPTO's "Eligibility Quick Reference Sheet Identifying Abstract Ideas," which has been superseded by this document.

In the rare circumstance in which an examiner believes a claim limitation that does not fall within the enumerated groupings of abstract ideas should nonetheless be treated as reciting an abstract idea, the procedure described in Section III.C for analyzing the claim should be followed.

For laws of nature and natural phenomena, Prong One does not represent a change. Examiners should continue to follow existing guidance to identify whether a claim recites one of these

GUARDANT HEALTH EX.1009.446

exceptions,[23] and if it does, proceed to Prong Two of the 2019 Revised Patent Subject Matter Eligibility Guidance in order to evaluate whether the claim integrates the law of nature or natural phenomenon into a practical application.

2. Prong Two: If The Claim Recites A Judicial Exception, Evaluate Whether The Judicial Exception Is Integrated Into A Practical Application

In Prong Two, examiners should evaluate whether the claim as a whole integrates the recited judicial exception into a practical application of the exception. A claim that integrates a judicial exception into a practical application will apply, rely on, or use the judicial exception in a manner that imposes a meaningful limit on the judicial exception, such that the claim is more than a drafting effort designed to monopolize the judicial exception. When the exception is so integrated, then the claim is not directed to a judicial exception (Step 2A: NO) and is eligible. This concludes the eligibility analysis. If the additional elements do not integrate the exception into a practical application, then the claim is directed to the judicial exception (Step 2A: YES), and requires further analysis under Step 2B (where it may still be eligible if it amounts to an inventive concept), as explained in Section III.B of the 2019 Revised Patent Subject Matter Eligibility Guidance.

Prong Two represents a change from prior guidance. The analysis under Prong Two is the same for all claims reciting a judicial exception, whether the exception is an abstract idea, a law of nature, or a natural phenomenon.

---

[23] *See* MPEP 2106.04(b)-(c).

GUARDANT HEALTH EX.1009.447

Examiners evaluate integration into a practical application by: (a) identifying whether there are any additional elements recited in the claim beyond the judicial exception(s); and (b) evaluating those additional elements individually and in combination to determine whether they integrate the exception into a practical application, using one or more of the considerations laid out by the Supreme Court and the Federal Circuit, for example those listed below. While some of the considerations listed below were discussed in prior guidance in the context of Step 2B, evaluating them in revised Step 2A promotes early and efficient resolution of patent eligibility, and increases certainty and reliability. Examiners should note, however, that revised Step 2A specifically excludes consideration of whether the additional elements represent well-understood, routine, conventional activity. Instead, analysis of well-understood, routine, conventional activity is done in Step 2B. Accordingly, in revised Step 2A examiners should ensure that they give weight to all additional elements, whether or not they are conventional, when evaluating whether a judicial exception has been integrated into a practical application.

In the context of revised Step 2A, the following exemplary considerations are indicative that an additional element (or combination of elements)[24] may have integrated the exception into a practical application:

- an additional element reflects an improvement in the functioning of a computer, or an improvement to other technology or technical field;[25]

[24] USPTO guidance uses the term "additional elements" to refer to claim features, limitations, and/or steps that are recited in the claim beyond the identified judicial exception. Again, whether an additional element or combination of elements integrate the exception into a practical application should be evaluated on the claim as a whole.

[25] For example, a modification of Internet hyperlink protocol to dynamically produce a dual-source hybrid webpage. See MPEP 2106.05(a) for more information concerning improvements in the functioning of a computer or to any other technology or technical field, including a discussion of the exemplar provided herein, which is based on *DDR Holdings*, 773 F.3d at 1258-59. See also USPTO *Finjan* Memorandum (discussing *Finjan* and *Core Wireless*).

GUARDANT HEALTH EX.1009.448

- an additional element that applies or uses a judicial exception to effect a particular treatment or prophylaxis for a disease or medical condition;[26]
- an additional element implements a judicial exception with, or uses a judicial exception in conjunction with, a particular machine or manufacture that is integral to the claim;[27]
- an additional element effects a transformation or reduction of a particular article to a different state or thing;[28] and
- an additional element applies or uses the judicial exception in some other meaningful way beyond generally linking the use of the judicial exception to a particular technological environment, such that the claim as a whole is more than a drafting effort designed to monopolize the exception.[29]

This is not an exclusive list, and there may be other examples of integrating the exception into a practical application.

---

[26] For example, an immunization step that integrates an abstract idea into a specific process of immunizing that lowers the risk that immunized patients will later develop chronic immune-mediated diseases. *See, e.g.*, *Classen Immunotherapies, Inc. v. Biogen IDEC*, 659 F.3d 1057, 1066-68 (Fed. Cir. 2011). *See also Vanda Pharm. Inc. v. West-Ward Pharm. Int'l Ltd.*, 887 F.3d 1117, 1135 (Fed. Cir. 2018) (holding claims to the practical application of the natural relationships between iloperidone, CYP2D6 metabolism, and QTc prolongation to treat schizophrenia, not merely the recognition of those relationships, to be patent eligible at *Mayo/Alice* step 1 (USPTO Step 2A)), and USPTO *Vanda* Memorandum (discussing *Vanda*).

[27] For example, a Fourdrinier machine (which is understood in the art to have a specific structure comprising a headbox, a paper-making wire, and a series of rolls) that is arranged in a particular way that uses gravity to optimize the speed of the machine while maintaining quality of the formed paper web. See MPEP 2106.05(b) for more information concerning use of a judicial exception with, or in conjunction with, a particular machine or manufacture, including a discussion of the exemplar provided herein, which is based on *Eibel Process Co. v. Minnesota & Ontario Paper Co.*, 261 U.S. 45, 64-65 (1923).

[28] For example, a process that transforms raw, uncured synthetic rubber into precision-molded synthetic rubber products by using a mathematical formula to control operation of the mold. See MPEP 2106.05(c) for more information concerning transformation or reduction of a particular article to a different state or thing, including a discussion of the exemplar provided herein, which is based on *Diehr*, 450 U.S. at 184.

[29] For example, a combination of steps including installing rubber in a press, closing the mold, constantly measuring the temperature in the mold, and automatically opening the press at the proper time, all of which together meaningfully limited the use of a mathematical equation to a practical application of molding rubber products. See MPEP 2106.05(e) for more information on this consideration, including a discussion of the exemplar provided herein, which is based on *Diehr*, 450 U.S. at 184, 187. See also USPTO *Finjan* Memorandum (discussing *Finjan* and *Core Wireless*).

GUARDANT HEALTH EX.1009.449

The courts have also identified examples in which a judicial exception has not been integrated into a practical application:

- an additional element merely recites the words "apply it" (or an equivalent) with the judicial exception, or merely includes instructions to implement an abstract idea on a computer, or merely uses a computer as a tool to perform an abstract idea;[30]
- an additional element adds insignificant extra-solution activity to the judicial exception;[31] and
- an additional element does no more than generally link the use of a judicial exception to a particular technological environment or field of use.[32]

It is critical that examiners consider the claim as a whole when evaluating whether the judicial exception is meaningfully limited by integration into a practical application of the exception. Some elements may be enough on their own to meaningfully limit an exception, but other times

[30] For example, a limitation indicating that a particular function such as creating and maintaining electronic records is performed by a computer, without specifying how. See MPEP 2106.05(f) for more information concerning mere instructions to apply a judicial exception, including a discussion of the exemplar provided herein, which is based on *Alice*, 573 U.S. at 222-26. *See also Benson*, 409 U.S. 63 (holding that merely implementing a mathematical principle on a general purpose computer is a patent ineligible abstract idea); *Credit Acceptance Corp. v. Westlake Services*, 859 F.3d 1044 (Fed. Cir. 2017) (using a computer as a tool to process an application for financing a purchase).

[31] For example, a mere data gathering such as a step of obtaining information about credit card transactions so that the information can be analyzed in order to detect whether the transactions were fraudulent. See MPEP 2106.05(g) for more information concerning insignificant extra-solution activity, including a discussion of the exemplar provided herein, which is based on *CyberSource*, 654 F.3d at 1375. *See also Mayo*, 566 U.S. at 79 (concluding that additional element of measuring metabolites of a drug administered to a patient was insignificant extra-solution activity, which was insufficient to confer patent eligibility); *Flook*, 437 U.S. at 590 (step of adjusting an alarm limit based on the output of a mathematical formula was "post-solution activity" and did not render method patent eligible).

[32] For example, a claim describing how the abstract idea of hedging could be used in the commodities and energy markets, or a claim limiting the use of a mathematical formula to the petrochemical and oil-refining fields. See MPEP 2106.05(h) concerning generally linking use of a judicial exception to a particular technological environment or field of use, including a discussion of the exemplars provided herein, which are based on *Bilski*, 561 U.S. at 612, and *Flook*, 437 U.S. at 588-90. Thus, the mere application of an abstract method of organizing human activity in a particular field is not sufficient to integrate the judicial exception into a practical application.

GUARDANT HEALTH EX.1009.450

it is the combination of elements that provide the practical application. When evaluating whether an element (or combination of elements) integrates an exception into a practical application, examiners should give careful consideration to both the element and how it is used or arranged in the claim as a whole. Because revised Step 2A does not evaluate whether an additional element is well-understood, routine, conventional activity, examiners are reminded that a claim that includes conventional elements may still integrate an exception into a practical application, thereby satisfying the subject matter eligibility requirement of Section 101.[33]

B. Step 2B: If The Claim Is Directed To A Judicial Exception, Evaluate Whether The Claim Provides An Inventive Concept

It is possible that a claim that does not "integrate" a recited judicial exception is nonetheless patent eligible. For example the claim may recite additional elements that render the claim patent eligible even though a judicial exception is recited in a separate claim element.[34] Along these lines, the Federal Circuit has held claims eligible at the second step of the *Alice/Mayo* test (USPTO Step 2B) because the additional elements recited in the claims provided "significantly more" than the recited judicial exception (e.g., because the additional elements were unconventional in combination).[35] Therefore, if a claim has been determined to be directed to a judicial exception under revised Step 2A, examiners should then evaluate the additional elements

---

[33] Of course, such claims must also satisfy the other conditions and requirements of patentability, for example, under section 102 (novelty), 103 (nonobviousness), and 112 (enablement, written description, definiteness). *Bilski*, 561 U.S. at 602.

[34] *See, e.g., Diehr*, 450 U.S. at 187 ("Our earlier opinions lend support to our present conclusion that a claim drawn to subject matter otherwise statutory does not become nonstatutory simply because it uses a mathematical formula, computer program, or digital computer."); *id.* at 185 ("Our conclusion regarding respondents' claims is not altered by the fact that in several steps of the process a mathematical equation and a programmed digital computer are used.").

[35] *See, e.g., Amdocs*, 841 F.3d at 1300, 1303; *BASCOM*, 827 F.3d at 1349-52; *DDR Holdings*, 773 F.3d at 1257-59.

GUARDANT HEALTH EX.1009.451

individually and in combination under Step 2B to determine whether they provide an inventive concept (i.e., whether the additional elements amount to significantly more than the exception itself). If the examiner determines that the element (or combination of elements) amounts to significantly more than the exception itself (Step 2B: YES), the claim is eligible, thereby concluding the eligibility analysis. If the examiner determines that the element and combination of elements does not amount to significantly more than the exception itself, the claim is ineligible (Step 2B: NO) and the examiner should reject the claim for lack of subject matter eligibility.

While many considerations in Step 2A need not be reevaluated in Step 2B, examiners should continue to consider in Step 2B whether an additional element or combination of elements:

- adds a specific limitation or combination of limitations that are not well-understood, routine, conventional activity in the field, which is indicative that an inventive concept may be present; or
- simply appends well-understood, routine, conventional activities previously known to the industry, specified at a high level of generality, to the judicial exception, which is indicative that an inventive concept may not be present.[36]

For this reason, if an examiner had previously concluded under revised Step 2A that, e.g., an additional element was insignificant extra-solution activity, they should reevaluate that conclusion in Step 2B. If such reevaluation indicates that the element is unconventional or

[36] In accordance with existing guidance, an examiner's conclusion that an additional element (or combination of elements) is well understood, routine, conventional activity must be supported with a factual determination. For more information concerning evaluation of well-understood, routine, conventional activity, see MPEP 2106.05(d), as modified by the USPTO *Berkheimer* Memorandum.

GUARDANT HEALTH EX.1009.452

otherwise more than what is well-understood, routine, conventional activity in the field, this finding may indicate that an inventive concept is present and that the claim is thus eligible.[37] For example, when evaluating a claim reciting an abstract idea such as a mathematical equation and a series of data gathering steps that collect a necessary input for the equation, an examiner might consider the data gathering steps to be insignificant extra-solution activity in revised Step 2A, and therefore find that the judicial exception is not integrated into a practical application.[38] However, when the examiner reconsiders the data gathering steps in Step 2B, the examiner could determine that the combination of steps gather data in an unconventional way and therefore include an "inventive concept," rendering the claim eligible at Step 2B.[39] Likewise, a claim that does not meaningfully integrate a judicial exception into a practical application of the exception sufficient to pass muster at Step 2A, may nonetheless include additional subject matter that is unconventional and thus an "inventive concept" at Step 2B.[40]

---

[37] *Mayo*, 566 U.S. at 82 ("[S]imply appending conventional steps, specified at a high level of generality, to laws of nature, natural phenomena, and abstract ideas cannot make those laws, phenomena, and ideas patentable."); *but see id.* at 85 ("[T]he claimed process included not only a law of nature but also several unconventional steps (such as inserting the receptacle, applying heat to the receptacle externally, and blowing the air into the furnace) that confined the claims to a particular, useful application of the principle." (discussing the old English case, *Neilson v. Harford*, Webster's Patent Cases 295 (1841))).

[38] *See supra* note 34; *see also OIP Techs.*, 788 F.3d at 1363 (finding that gathering statistics generated based on customer testing for input to a pricing calculation "fail[s] to 'transform' the claimed abstract idea into a patent-eligible invention").

[39] *Compare Flook*, 437 U.S. at 585–86 (holding claimed method of updating alarm limits to be ineligible because: "In essence, the method consists of three steps: an initial step which merely measures the present value of the process variable (e.g., the temperature); an intermediate step which uses an algorithm to calculate an updated alarm-limit value; and a final step in which the actual alarm limit is adjusted to the updated value. The only difference between the conventional methods of changing alarm limits and that described in respondent's application rests in the second step—the mathematical algorithm or formula."); *with Exergen Corp. v. Kaz USA, Inc.*, 725 F. App'x 959, 966 (Fed. Cir. 2018) (holding claimed body temperature detector to be eligible because: "Here, the patent is directed to the measurement of a natural phenomenon (core body temperature). Even if the concept of such measurement is directed to a natural phenomenon and is abstract at step one, the measurement method here was not conventional, routine, and well-understood. Following years and millions of dollars of testing and development, the inventor determined for the first time the coefficient representing the relationship between temporal-arterial temperature and core body temperature and incorporated that discovery into an unconventional method of temperature measurement.").

[40] *Compare Berkheimer*, 881 F.3d at 1370 (holding independent claim 1 to be ineligible at *Alice* step 2: "The[] conventional limitations of claim 1, combined with limitations of analyzing and comparing data and reconciling

GUARDANT HEALTH EX.1009.453

C. Treating a Claim Limitation That Does Not Fall Within the Enumerated Groupings of Abstract Ideas as Reciting an Abstract Idea

In the rare circumstance in which an examiner believes a claim limitation that does not fall within the enumerated groupings of abstract ideas should nonetheless be treated as reciting an abstract idea ("tentative abstract idea"), the examiner should evaluate whether the claim as a whole integrates the recited tentative abstract idea into a practical application as explained in Section III.A.2. If the claim as a whole integrates the recited tentative abstract idea into a practical application, the claim is not directed to a judicial exception (Step 2A: NO) and is eligible (thus concluding the eligibility analysis). If the claim as a whole does not integrate the recited tentative abstract idea into a practical application, then the examiner should evaluate the additional elements individually and in combination to determine whether they provide an inventive concept as explained in Section III.B. If an additional element or combination of additional elements provides an inventive concept as explained in Section III.B (Step 2B: YES), the claim is eligible (thus concluding the eligibility analysis). If the additional element or

---

differences between the data, fail to transform the abstract idea into a patent-eligible invention. The limitations amount to no more than performing the abstract idea of parsing and comparing data with conventional computer components") (internal quotation marks and citation omitted); *with id.* (concluding that dependent claims 4-7 may be eligible: "Claims 4–7, in contrast, contain limitations directed to the arguably unconventional inventive concept described in the specification. Claim 4 recites 'storing a reconciled object structure in the archive without substantial redundancy.' The specification states that storing object structures in the archive without substantial redundancy improves system operating efficiency and reduces storage costs. It also states that known asset management systems did not archive documents in this manner. Claim 5 depends on claim 4 and further recites 'selectively editing an object structure, linked to other structures to thereby effect a one-to-many change in a plurality of archived items.' The specification states one-to-many editing substantially reduces effort needed to update files because a single edit can update every document in the archive linked to that object structure. This one-to-many functionality is more than 'editing data in a straightforward copy-and-paste fashion,' as characterized by the district court. According to the specification, conventional digital asset management systems cannot perform one-to-many editing because they store documents with numerous instances of redundant elements, rather than eliminate redundancies through the storage of linked object structures. Claims 6–7 depend from claim 5 and accordingly contain the same limitations. These claims recite a specific method of archiving that, according to the specification, provides benefits that improve computer functionality. . . . [T]here is at least a genuine issue of material fact in light of the specification regarding whether claims 4–7 archive documents in an inventive manner that improves these aspects of the disclosed archival system.") (internal quotation marks and citations omitted).

GUARDANT HEALTH EX.1009.454

combination of additional elements does not provide an inventive concept as explained in Section III.B (Step 2B: NO), the examiner should bring the application to the attention of the Technology Center Director. Any rejection in which a claim limitation, which does not fall within the enumerated abstract ideas (tentative abstract idea), is nonetheless treated as reciting an abstract idea must be approved by the Technology Center Director (which approval will be indicated in the file record of the application), and must provide a justification[41] for why such claim limitation is being treated as reciting an abstract idea.[42]

D. Compact Prosecution

Regardless of whether a rejection under 35 U.S.C. 101 is made, a complete examination should be made for every claim under each of the other patentability requirements: 35 U.S.C. 102, 103, 112, and 101 (utility, inventorship and double patenting) and non-statutory double patenting.[43] Compact prosecution, however, does not mandate that the patentability requirements be analyzed in any particular order.

Dated: December 20, 2018.

Andrei Iancu,
Under Secretary of Commerce for Intellectual Property and
Director of the United States Patent and Trademark Office.

---

[41] Such justification may include, for example, an explanation of why the element contains subject matter that, per se, invokes eligibility concerns similar to those expressed by the Supreme Court with regard to the judicial exceptions. *See supra* note 5.

[42] Similarly, in the rare circumstance in which a panel of administrative patent judges (or panel majority) believes that a claim reciting a tentative abstract idea should be treated as reciting an abstract idea, the matter should be brought to the attention of the PTAB leadership by a written request for clearance.

[43] *See* MPEP 2103 et seq. and 2106(III).

GUARDANT HEALTH EX.1009.455

[FR Doc. 2018-28282 Filed: 1/4/2019 8:45 am; Publication Date: 1/7/2019]

GUARDANT HEALTH EX.1009.456

REMARKS

Claims 68-87 are pending in the subject application with claim 76 withdrawn by the Examiner. By this Amendment, applicants have hereinabove amended claims 68, 69, 75, 76, 77, 81, 85, and 87. Support for the amended claims may be found, *inter alia*, in the specification as filed as shown in the table below:

Claim	Specification
68	Page 15, lines 2-26; page 44, lines 15-21; page 51, lines 9-11; page 45, lines 11-14 and 28-33
69	Page 15, line 27 to page 16, line 2
75	Page 18, line 18 to page 19, line 29,;page 44, lines 15-21; page 51, lines 9-11; page 45, lines 11-14 and 28-33
76	Page 20, line 1 to page 21 line 7
77	Page 21, line 8 to page 22, line 3
81	Page 24, lines 11-14
85	Page 29, lines 4-6
87	Page 29, lines 4-6

The claim amendments herein are appropriate for entry under 37 C.F.R. § 1.116 because the amendments (1) comply with requirements expressly set forth in the November 30, 2018 Final Office Action, and (2) do not raise new issues that would require further consideration and/or search. Specifically, the applicants have amended the claims to address the Examiner's claim objections and claims rejections under 35 U.S.C. §112, second paragraph, set forth in the November 30, 2018 Office Action. Therefore, the amendments are proper and should be entered.

Additionally, the claim amendments herein are appropriate for consideration under After Final Consideration Pilot 2.0 because they contain an amendment to at least one independent claim that does not broaden the scope of the independent claim in any aspect. Specifically, claims 68 and 75 have been amended to further specify

that "each count is an estimate of the relative genomic copy number for each location of the genome" and that the method comprises "obtaining more than one count to obtain counts."

Further, in consideration of the amendments herein and in view of the applicants remarks hereinbelow, applicants submit that claims 68-76 and 78-87 are allowable. In the January 18, 2018 Restriction Requirement issued in connection with the above-identified application, the Examiner required applicants to elect a species from among two species as identified by the Examiner, one directed to claim 76 and one directed to claim 77. In applicants Response filed March 16, 2018, applicants elected the species directed to claim 76 as identified by the Examiner. In the January 18, 2018 Restriction Requirement, the Examiner further indicated that upon allowance of a generic claim, applicants will be entitled to consideration of claims to additional species which depend from or otherwise require all the limitations of an allowable generic claim as provided by 37 C.F.R. 1.141. The Examiner identified claims 68-75 and 78-87 as generic. Applicant has amended claim 76 in view of the Examiner's rejections to claim 77. Accordingly, applicants request rejoinder of claim 77.

Accordingly, upon entry of this Amendment, claims 68-87 are pending and under consideration in the subject application.

Objections to the Drawings

In the November 30, 2018 Final Office Action, the Examiner objected to Figures 7-10 for allegedly containing letters that were difficult to recognize.

Applicants' reply

In response, applicants attach hereto as **Exhibit 1** EIGHT (8) replacement sheets of color drawings containing Figures 7-10.

Applicants submit that the letters in replacement Figures 7-10 are clear.

A Petition Under 37 C.F.R. §1.84(a)(2) to Accept Colored Figures Filed in Connection with the Above-Identified Application was filed on March 7, 2016 along with payment of the corresponding fee under 37 C.F.R. §1.84(a)(2)(1).

Applicants submit that such drawings are necessary because certain features of the Figures would not be identifiable or distinguishable in black and white.

In accordance with the guidance set forth in Section H of "Legal Framework for Electronic Filing System-Web (EFS-Web)" [Federal Register 74:206 (October 27, 2009) pp. 55200-55211], the requirement for three (3) sets of color drawings or figures under 37 C.F.R. §1.84(a)(2)(ii) is not applicable to color drawings or figures submitted via EFS-Web. Therefore, applicants are submitting via EFS-Web only one (1) set of figures.

In accordance with 37 C.F.R. §1.84(a)(2)(iii), the subject specification contains a paragraph immediately after the section heading titled "Brief Description Of The Figures" stating:

> The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application with color drawing(s) will be provided by the Office upon request and payment of the necessary fee.

Applicants maintain that this Petition complies with the requirements of 37 C.F.R. §1.84(a)(2), and respectfully request that this Petition be granted and the sheets of colored figures filed with the subject application be accepted.

Applicants maintain that amendments to the drawings and the specification raise no issue of new matter.

Accordingly, applicants respectfully request that the objections to the drawings be withdrawn.

Objections to the Claims

In the November 30, 2018 Final Office Action, the Examiner objected to claims 68, 69, 77, 81, and 85 or 87. The Examiner's rationale and suggested claim amendments may be found on pages 2 and 3 of the November 30, 2018 Office Action.

Applicants' reply

In response, without conceding correctness of the Examiner's position, and solely for the purpose of advancing prosecution, applicants have amended claims 68, 69, 77, 81, 85, and 87.

Applicant has amended claims 77, 81, 85, and 87 in accordance with the Examiner's suggested language.

In Section 3 of the November 30, 2018 Final Office Action the Examiner suggested amending claim 68 to recite "and obtaining genomic copy number information from the genomic nucleic acids." In Section 10 of the November 30, 2018 Final Office Action the Examiner requested additional clarification of step (f). To address both issues, applicants have amended claim 68 to further specify that "wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome" Accordingly, applicant submits that Claim 68 as amended is allowable in view of the Examiner's objection.

In Section 4 of the November 30, 2018 Final Office Action the Examiner

suggested amending claim 69 to remove the recitation of "within a region of the genome." In response, applicant has amended claim 69 to remove the recitation of "on the genome." Applicant submits that specification makes clear that "the term 'region of the genome' refers to a continuous genomic sequence comprising multiple discrete locations.'" (Specification at page 36, lines 17-19). Accordingly, applicant submits that Claim 69 as amended is allowable in view of the Examiner's objection.

Accordingly, applicants respectfully request that the claim objections be withdrawn.

Rejection of claims under 35 U.S.C. § 112, Second Paragraph

In the November 30, 2018 Final Office Action the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 112, second paragraph, as allegedly indefinite for failing to particularly point out and distinctly claim the subject matter which applicants regard as the invention. The Examiner's rationale is set forth on pages 3 and 4 of the November 30, 2018 Office Action.

Applicants' reply

As an initial matter, without conceding correctness of the Examiner's position, and solely for the purpose of advancing prosecution, applicants have amended claims 68 and 75.

Specifically, claims 68 and 75 have been amended to further specify that "wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome".

Accordingly, applicants submit that claims 68 and 75 as amended clarify how copy number is obtained from the amplified tagged nucleic

acid molecules. Further, claim 68 now recites the term "counts," providing antecedent basis for this limitation in claims 70, 71, 72, and 74.

Accordingly, applicants respectfully request that the claim rejection under 35 U.S.C. § 112, second paragraph be withdrawn.

Rejection of claims under 35 U.S.C. § 101

In the November 30, 2018 Final Office Action the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 101, as allegedly not directed to patent eligible subject matter. The Examiner's rationale is set forth on pages 4-9 of the November 30, 2018 Final Office Action.

Applicants' reply

In response, applicants respectfully traverse the Examiner's rejection.

Applicants emphasize that the United States Patent and Trademark Office issued "2019 Revised Patent Subject Matter Eligibility Guidance" on January 7, 2019, attached hereto as **Exhibit 2**, (hereinafter the "Revised Guidance") which is intended to assist Examiners in evaluating subject matter eligibility.

The Revised Guidance clarifies that "[o]nly when a claim recites a judicial exception and fails to integrate the exception into a practical application, is the claim 'directed' to a judicial exception." (Revised Guidance at page 51, emphasis added). The Revised Guidance further clarifies that this "adds a more detailed eligibility analysis to step one of the *Alice/Mayo* test (USPTO Step 2A) than was required under prior guidance." (Revised Guidance at page 54).

The Revised Guidance further advises that:

> Examiners evaluate integration into a practical application by: (a) identifying whether there are any additional elements recited in the claim beyond the judicial exception(s); and (b) evaluating those additional elements individually and in combination to determine whether they integrate the exception into a practical application, using one or more of the considerations laid out by the Supreme Court and the Federal Circuit, for example those listed below.

(Revised Guidance at pages 54 and 55). Among the examples listed, the Revised Guidance lists "an additional element [that] effects a transformation or reduction of a particular article to a different state or thing." (Revised Guidance at page 55).

With regard to these additional elements, the Revised Guidance further instructs that:

> Examiner's should note, however, that revised Step 2A specifically excludes consideration of whether the additional elements represent well-understood, routine, conventional activity. Instead, analysis of well-understood, routine, conventional activity is done in Step 2B. Accordingly, in revised Step 2A examiners should ensure that they give weight to all additional elements, whether or not they are conventional, when evaluating whether a judicial exception has been integrated into a practical application.

(Revised Guidance at page 55).

Regarding this guidance, the pending claims are directed to a method of sequencing that (i) comprises specific steps (specific elements) (a)-(c) that transform genomic nucleic acids to generate new, non-naturally occurring molecules (unique tagged nucleic acid molecules) that are sequenced and mapped to a location on the genome, and then (ii) a step to count the different, mapped, transformed molecules for the practical purpose (application) of obtaining genomic copy number information, for example, to generate a genomic profile of a cell (for example a cancer cell) or of a population of cells. In this way, the

specific transformation steps (elements) required by the claimed method integrate the mapping and counting steps into a practical application, in a manner that avoids distortion of copy number by amplification of the genomic nucleic acids.

Accordingly, in light of the Revised Guidance, applicants submit that the pending claims comply with 35 U.S.C. § 101 and respectfully request that this rejection be withdrawn.

Non-statutory Double Patenting

In the November 30, 2018 Final Office Action, the Examiner rejected claims 68-75 and 77-87 on the grounds of nonstatutory double patenting (ODP), as allegedly unpatentable over claims 1-20 of U.S. Patent No. 9,404,156 B2.

Applicants' Reply

In response, without conceding correctness of the Examiner's position, and solely for the purpose of advancing prosecution, applicants submit herewith, as **Exhibit 3**, a Terminal Disclaimer signed by an authorized official of the assignee of record of U.S. Patent No. 9,404,156 and of the subject application, namely Cold Spring Harbor Laboratory.

Under 37 C.F.R. § 1.321(c), a Terminal Disclaimer must be accompanied by the fee set forth in 37 C.F.R. § 1.20(d). Under 37 C.F.R. § 1.20(d), the fee for filing a Terminal Disclaimer is ONE HUNDRED SIXTY DOLLARS ($160.00). Accordingly, authorization is hereby given to charge this amount of fee to Deposit Account No. 03-3125.

In accordance with 37 C.F.R. § 1.321(c), the Terminal Disclaimer submitted herewith as **Exhibit 3**: (1) is signed by the assignee of record of the entire interest of the subject application, (2) specifies the portion of the term of patent being disclaimed, (3) states the present extent of assignee's ownership interest in the

patent to be granted, (4) and is accompanied by the fee set forth in 37 C.F.R. § 1.20(d). Accordingly, the Terminal Disclaimer submitted herewith complies with the requirements of 37 C.F.R. § 1.321(c).

Additionally, applicant attaches hereto as **Exhibit 4** a statement under 37 C.F.R. §3.73(c). Both the Statement under 37 C.F.R. §3.73(c) and the Terminal Disclaimer establish that Cold Spring Harbor Laboratory is the assignee of the entire right, title, and interest in the above-identified application pursuant to 37 C.F.R. §3.73(c). As assignee of the above-identified application, Cold Spring Harbor Laboratory is entitled to conduct prosecution of the subject application pursuant to §3.71.

Based on the submission of the Terminal Disclaimer, the double patenting rejections set forth in the November 30, 2018 Office Action have been overcome.

Accordingly, applicants respectfully request withdrawal of this rejection.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the $160.00 fee for the submission of a Terminal Disclaimer and the $300.00 fee for a Petition for a Two-month Extension of Time for a small entity, is deemed necessary in connection with the filing of this Amendment. However, if any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

2019-04-17
Adftya Ramanathan Date

Docket No. 5981/81503-AA/GJG/ADR

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112

BY EFS
Commissioner for Patents
Alexandria, VA

TERMINAL DISCLAIMER

Cold Spring Harbor Laboratory, One Bungtown Road, Cold Spring Harbor, New York 11724, U.S.A., is the owner of (assignee of record of the entire right, title and interest in) the above-identified application by virtue of an assignment of U.S. Serial No. 13/278,333, filed October 21, 2011, the parent of the above-identified application, which assignment was recorded with the U.S. Patent and Trademark Office in connection with U.S. Serial No. 13/278,333 on January 16, 2012 at Reel No. 027537, Frame 0950-0955. A copy of the recorded assignment is attached hereto as **Exhibit A**. The subject application, U.S. Serial No. 15/063,278 is a continuation of U.S. Serial No. 13/278,333. Since the Assignment of rights attached hereto as **Exhibit A** refers to U.S. Serial No. 13/278,333 and continuing applications therefrom, each of U.S. Serial No. 13/278,333 and the above-identified application are owned by Cold Spring Harbor Laboratory.

Cold Spring Harbor Laboratory is also the owner of (assignee of record of the entire right, title and interest in) U.S. Patent No. 9,404,156 issued August 2, 2016, from aforementioned U.S. Serial No. 13/278,333 by virtue of the document attached hereto as **Exhibit A**.

GUARDANT HEALTH EX.1009.467

Cold Spring Harbor Laboratory hereby disclaims, except as provided below, the terminal part of the term of any patent granted on the above-identified application which would extend beyond the expiration date of the full statutory term of U.S. Patent No. 9,404,156, and hereby agrees that any patent issued from the subject application shall be enforceable only for and during such period that the owner of such patent is the same as the owner of U.S. Patent No. 9,404,156, this agreement to run with any patent granted on the subject application and to be binding upon the grantee, its successors and assigns.

In making the above disclaimer, Cold Spring Harbor Laboratory does not disclaim the terminal part of the term of any patent granted on the subject application that would extend to the expiration date of the full statutory term of U.S. Patent No. 9,404,156 in the event that such earliest to expire patent expires prior to its full statutory term for failure to pay a maintenance fee, is held unenforceable, is found invalid by a court of competent jurisdiction, is statutorily disclaimed in whole or terminally disclaimed under 37 C.F.R. §1.321, has all claims canceled by a reexamination certificate, is reissued, or is in any other manner terminated prior to the expiration date of the full statutory term to which such earliest to expire patent would otherwise have been entitled.

I have reviewed the recorded Assignment document attached hereto as **Exhibit A** and certify, to the best of my knowledge and belief, that Cold Spring Harbor Laboratory is the assignee of all right, title and interest in and to the subject application and U.S. Patent No. 9,404,156. I further certify that I am authorized to sign this Terminal Disclaimer on behalf of Cold Spring Harbor Laboratory.

I hereby declare that all statements made herein of my own knowledge are true, and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that any such willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section

1001 of Title 18 of the United States Code and that any such willful false statement any jeopardize the validity of this application or any patent issued thereon.

Date: 4/17/2019 Cold Spring Harbor Laboratory

By: _____

Vladimir Drozdov, Ph.D., J.D.
Chief Patent Counsel
Office of Technology Transfer
Cold Spring Harbor Laboratory

PTO/AIA/96 (08-12)
Approved for use through 01/31/2013. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

STATEMENT UNDER 37 CFR 3.73(c)

Applicant/Patent Owner: Cold Spring Harbor Laboratory

Application No./Patent No.: 15/063,278 Filed/Issue Date: March 7, 2016

Titled: Varietal Counting of Nucleic Acids for Obtaining Genomic Copy Number Information

Cold Spring Harbor Laboratory, a corporation

(Name of Assignee) (Type of Assignee, e.g., corporation, partnership, university, government agency, etc.)

states that, for the patent application/patent identified above, it is (choose **one** of options 1, 2, 3 or 4 below):

1. ☑ The assignee of the entire right, title, and interest.

2. ☐ An assignee of less than the entire right, title, and interest (check applicable box):

   ☐ The extent (by percentage) of its ownership interest is _____%. Additional Statement(s) by the owners holding the balance of the interest must be submitted to account for 100% of the ownership interest.

   ☐ There are unspecified percentages of ownership. The other parties, including inventors, who together own the entire right, title and interest are:

   Additional Statement(s) by the owner(s) holding the balance of the interest must be submitted to account for the entire right, title, and interest.

3. ☐ The assignee of an undivided interest in the entirety (a complete assignment from one of the joint inventors was made). The other parties, including inventors, who together own the entire right, title, and interest are:

   Additional Statement(s) by the owner(s) holding the balance of the interest must be submitted to account for the entire right, title, and interest.

4. ☐ The recipient, via a court proceeding or the like (*e.g.*, bankruptcy, probate), of an undivided interest in the entirety (a complete transfer of ownership interest was made). The certified document(s) showing the transfer is attached.

The interest identified in option 1, 2 or 3 above (not option 4) is evidenced by either (choose **one** of options A or B below):

A. ☑ An assignment from the inventor(s) of the patent application/patent identified above. The assignment was recorded in the United States Patent and Trademark Office at Reel 027537, Frame 0950/0955, or for which a copy thereof is attached.

B. ☐ A chain of title from the inventor(s), of the patent application/patent identified above, to the current assignee as follows:

   1. From: _____ To: _____

      The document was recorded in the United States Patent and Trademark Office at

      Reel _____, Frame _____, or for which a copy thereof is attached.

   2. From: _____ To: _____

      The document was recorded in the United States Patent and Trademark Office at

      Reel _____, Frame _____, or for which a copy thereof is attached.

This collection of information is required by 37 CFR 3.73(b). The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199

PTO/AIA/96 (08-12)
Approved for use through 01/31/2013. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

STATEMENT UNDER 37 CFR 3.73(c)

3. From: _____ To: _____

The document was recorded in the United States Patent and Trademark Office at

Reel _____, Frame _____, or for which a copy thereof is attached.

4. From: _____ To: _____

The document was recorded in the United States Patent and Trademark Office at

Reel _____, Frame _____, or for which a copy thereof is attached.

5. From: _____ To: _____

The document was recorded in the United States Patent and Trademark Office at

Reel _____, Frame _____, or for which a copy thereof is attached.

6. From: _____ To: _____

The document was recorded in the United States Patent and Trademark Office at

Reel _____, Frame _____, or for which a copy thereof is attached.

☐ Additional documents in the chain of title are listed on a supplemental sheet(s).

☐ As required by 37 CFR 3.73(c)(1)(i), the documentary evidence of the chain of title from the original owner to the assignee was, or concurrently is being, submitted for recordation pursuant to 37 CFR 3.11.

[NOTE: A separate copy (i.e., a true copy of the original assignment document(s)) must be submitted to Assignment Division in accordance with 37 CFR Part 3, to record the assignment in the records of the USPTO. See MPEP 302.08]

The undersigned (whose title is supplied below) is authorized to act on behalf of the assignee.

Gary J. Gershik — Signature

4/17/19 — Date

Gary J. Gershik — Printed or Typed Name

39,992 — Title or Registration Number

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

The application data sheet is part of the provisional or nonprovisional application for which it is being submitted. The following form contains the bibliographic data arranged in a format specified by the United States Patent and Trademark Office as outlined in 37 CFR 1.76.
This document may be completed electronically and submitted to the Office in electronic format using the Electronic Filing System (EFS) or the document may be printed and included in a paper filed application.

Secrecy Order 37 CFR 5.2:

☐ Portions or all of the application associated with this Application Data Sheet may fall under a Secrecy Order pursuant to 37 CFR 5.2 (Paper filers only. Applications that fall under Secrecy Order may not be filed electronically.)

Inventor Information:

Inventor 1 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	James		Hicks	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	Locust Valley	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	3 Great Meadows Road		
Address 2			
City	Locust Valley	State/Province	NY
Postal Code	11560	Country	US

Inventor 2 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Nicholas		Navin	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	Houston	State/Province	TX	Country of Residence	US

Mailing Address of Inventor:

Address 1	1621 Wichita Street		
Address 2			
City	Houston	State/Province	TX
Postal Code	77004	Country	US

Inventor 3 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Jennifer		Troge	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

U.S. Serial No.: 15/063,278
Applicant(s): James Hicks et al.
Filed: March 7, 2016
Exhibit 4

GUARDANT HEALTH EX.1009.473

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

City	Oyster Bay	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	100 Simcoe Street
Address 2	

City	Oyster Bay	State/Province	NY
Postal Code	11771	Country	US

Inventor 4 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Zihua		Wang	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	Huntington	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	13 Meadowlawn Street
Address 2	

City	Huntington	State/Province	NY
Postal Code	11743	Country	US

Inventor 5 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Michael		Wigler	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	Cold Spring Harbor	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	52 Snake Hill Road
Address 2	

City	Cold Spring Harbor	State/Province	NY
Postal Code	11724	Country	US

All Inventors Must Be Listed - Additional Inventor Information blocks may be generated within this form by selecting the **Add** button. Add

Correspondence Information:

Enter either Customer Number or complete the Correspondence Information section below.
For further information see 37 CFR 1.33(a).

☐ **An Address is being provided for the correspondence Information of this application.**

GUARDANT HEALTH EX.1009.474

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Customer Number	23432		
Email Address	patentdocketing@cooperdunham.com	Add Email	Remove Email

Application Information:

Title of the Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION		
Attorney Docket Number	81503-AA/GJG/AWG/AC	Small Entity Status Claimed	☒
Application Type	Nonprovisional		
Subject Matter	Utility		
Total Number of Drawing Sheets (if any)	~~11~~ 15	Suggested Figure for Publication (if any)	

Filing By Reference:

Only complete this section when filing an application by reference under 35 U.S.C. 111(c) and 37 CFR 1.57(a). Do not complete this section if application papers including a specification and any drawings are being filed. Any domestic benefit or foreign priority information must be provided in the appropriate section(s) below (i.e., "Domestic Benefit/National Stage Information" and "Foreign Priority Information").

For the purposes of a filing date under 37 CFR 1.53(b), the description and any drawings of the present application are replaced by this reference to the previously filed application, subject to conditions and requirements of 37 CFR 1.57(a).

Application number of the previously filed application	Filing date (YYYY-MM-DD)	Intellectual Property Authority or Country

Publication Information:

☐ Request Early Publication (Fee required at time of Request 37 CFR 1.219)

☐ **Request Not to Publish.** I hereby request that the attached application not be published under 35 U.S.C. 122(b) and certify that the invention disclosed in the attached application **has not and will not** be the subject of an application filed in another country, or under a multilateral international agreement, that requires publication at eighteen months after filing.

Representative Information:

Representative information should be provided for all practitioners having a power of attorney in the application. Providing this information in the Application Data Sheet does not constitute a power of attorney in the application (see 37 CFR 1.32).
Either enter Customer Number or complete the Representative Name section below. If both sections are completed the customer Number will be used for the Representative Information during processing.

Please Select One:	◉ Customer Number	○ US Patent Practitioner	○ Limited Recognition (37 CFR 11.9)
Customer Number	23432		

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Domestic Benefit/National Stage Information:

This section allows for the applicant to either claim benefit under 35 U.S.C. 119(e), 120, 121, 365(c), or 386(c) or indicate National Stage entry from a PCT application. Providing benefit claim information in the Application Data Sheet constitutes the specific reference required by 35 U.S.C. 119(e) or 120, and 37 CFR 1.78.
When referring to the current application, please leave the "Application Number" field blank.

Prior Application Status	Pending		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
	Continuation of	13/278333	2011-10-21
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/510579	2011-07-22
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/406067	2010-10-22

Additional Domestic Benefit/National Stage Data may be generated within this form by selecting the **Add** button.

Foreign Priority Information:

This section allows for the applicant to claim priority to a foreign application. Providing this information in the application data sheet constitutes the claim for priority as required by 35 U.S.C. 119(b) and 37 CFR 1.55. When priority is claimed to a foreign application that is eligible for retrieval under the priority document exchange program (PDX)[i] the information will be used by the Office to automatically attempt retrieval pursuant to 37 CFR 1.55(i)(1) and (2). Under the PDX program, applicant bears the ultimate responsibility for ensuring that a copy of the foreign application is received by the Office from the participating foreign intellectual property office, or a certified copy of the foreign priority application is filed, within the time period specified in 37 CFR 1.55(g)(1).

			Remove
Application Number	Country[i]	Filing Date (YYYY-MM-DD)	Access Code[i] (if applicable)

Additional Foreign Priority Data may be generated within this form by selecting the **Add** button.

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

☐ This application (1) claims priority to or the benefit of an application filed before March 16, 2013 and (2) also contains, or contained at any time, a claim to a claimed invention that has an effective filing date on or after March 16, 2013.
NOTE: By providing this statement under 37 CFR 1.55 or 1.78, this application, with a filing date on or after March 16, 2013, will be examined under the first inventor to file provisions of the AIA.

GUARDANT HEALTH EX.1009.477

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Authorization or Opt-Out of Authorization to Permit Access:

When this Application Data Sheet is properly signed and filed with the application, applicant has provided written authority to permit a participating foreign intellectual property (IP) office access to the instant application-as-filed (see paragraph A in subsection 1 below) and the European Patent Office (EPO) access to any search results from the instant application (see paragraph B in subsection 1 below).

Should applicant choose not to provide an authorization identified in subsection 1 below, applicant **must opt-out** of the authorization by checking the corresponding box A or B or both in subsection 2 below.

NOTE: This section of the Application Data Sheet is **ONLY** reviewed and processed with the **INITIAL** filing of an application. After the initial filing of an application, an Application Data Sheet cannot be used to provide or rescind authorization for access by a foreign IP office(s). Instead, Form PTO/SB/39 or PTO/SB/69 must be used as appropriate.

1. Authorization to Permit Access by a Foreign Intellectual Property Office(s)

A. Priority Document Exchange (PDX) - Unless box A in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the European Patent Office (EPO), the Japan Patent Office (JPO), the Korean Intellectual Property Office (KIPO), the State Intellectual Property Office of the People's Republic of China (SIPO), the World Intellectual Property Organization (WIPO), and any other foreign intellectual property office participating with the USPTO in a bilateral or multilateral priority document exchange agreement in which a foreign application claiming priority to the instant patent application is filed, access to: (1) the instant patent application-as-filed and its related bibliographic data, (2) any foreign or domestic application to which priority or benefit is claimed by the instant application and its related bibliographic data, and (3) the date of filing of this Authorization. See 37 CFR 1.14(h)(1).

B. Search Results from U.S. Application to EPO - Unless box B in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the EPO access to the bibliographic data and search results from the instant patent application when a European patent application claiming priority to the instant patent application is filed. See 37 CFR 1.14(h)(2).

The applicant is reminded that the EPO's Rule 141(1) EPC (European Patent Convention) requires applicants to submit a copy of search results from the instant application without delay in a European patent application that claims priority to the instant application.

2. Opt-Out of Authorizations to Permit Access by a Foreign Intellectual Property Office(s)

☐ A. Applicant **DOES NOT** authorize the USPTO to permit a participating foreign IP office access to the instant application-as-filed. If this box is checked, the USPTO will not be providing a participating foreign IP office with any documents and information identified in subsection 1A above.

☐ B. Applicant **DOES NOT** authorize the USPTO to transmit to the EPO any search results from the instant patent application. If this box is checked, the USPTO will not be providing the EPO with search results from the instant application.

NOTE: Once the application has published or is otherwise publicly available, the USPTO may provide access to the application in accordance with 37 CFR 1.14.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Applicant Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

Applicant 1

If the applicant is the inventor (or the remaining joint inventor or inventors under 37 CFR 1.45), this section should not be completed. The information to be provided in this section is the name and address of the legal representative who is the applicant under 37 CFR 1.43; or the name and address of the assignee, person to whom the inventor is under an obligation to assign the invention, or person who otherwise shows sufficient proprietary interest in the matter who is the applicant under 37 CFR 1.46. If the applicant is an applicant under 37 CFR 1.46 (assignee, person to whom the inventor is obligated to assign, or person who otherwise shows sufficient proprietary interest) together with one or more joint inventors, then the joint inventor or inventors who are also the applicant should be identified in this section.

Clear

◯ Assignee	◯ Legal Representative under 35 U.S.C. 117	◯ Joint Inventor
◯ Person to whom the inventor is obligated to assign.	◯ Person who shows sufficient proprietary interest	

If applicant is the legal representative, indicate the authority to file the patent application, the inventor is:

Name of the Deceased or Legally Incapacitated Inventor:

If the Applicant is an Organization check here. ☒

Organization Name	Cold Spring Harbor Laboratory		
Mailing Address Information For Applicant:			
Address 1	One Bungtown Road		
Address 2			
City	Cold Spring Harbor	**State/Province**	NY
Country	US	Postal Code	11724
Phone Number		Fax Number	
Email Address			

Additional Applicant Data may be generated within this form by selecting the Add button.

Assignee Information including Non-Applicant Assignee Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

GUARDANT HEALTH EX.1009.479

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Assignee 1

Complete this section if assignee information, including non-applicant assignee information, is desired to be included on the patent application publication. An assignee-applicant identified in the "Applicant Information" section will appear on the patent application publication as an applicant. For an assignee-applicant, complete this section only if identification as an assignee is also desired on the patent application publication.

If the Assignee or Non-Applicant Assignee is an Organization check here. ☒

Organization Name	Cold Spring Harbor Laboratory

Mailing Address Information For Assignee including Non-Applicant Assignee:

Address 1	One Bungtown Road		
Address 2			
City	Cold Spring Harbor	**State/Province**	NY
Country i	US	Postal Code	11724
Phone Number		Fax Number	
Email Address			

Additional Assignee or Non-Applicant Assignee Data may be generated within this form by selecting the Add button.

Signature:

NOTE: This Application Data Sheet must be signed in accordance with 37 CFR 1.33(b). **However, if this Application Data Sheet is submitted with the INITIAL filing of the application and either box A or B is not checked in subsection 2 of the "Authorization or Opt-Out of Authorization to Permit Access" section, then this form must also be signed in accordance with 37 CFR 1.14(c).**

This Application Data Sheet **must** be signed by a patent practitioner if one or more of the applicants is a **juristic entity** (e.g., corporation or association). If the applicant is two or more joint inventors, this form must be signed by a patent practitioner, **all** joint inventors who are the applicant, or one or more joint inventor-applicants who have been given power of attorney (e.g., see USPTO Form PTO/AIA/81) on behalf of **all** joint inventor-applicants.

See 37 CFR 1.4(d) for the manner of making signatures and certifications.

Signature	/Gary J. Gershik/			Date (YYYY-MM-DD)	2019-04-17 ~~2018-03-07~~
First Name	Gary	Last Name	Gershik	Registration Number	39992

Additional Signature may be generated within this form by selecting the Add button.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

This collection of information is required by 37 CFR 1.76. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 23 minutes to complete, including gathering, preparing, and submitting the completed application data sheet form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

GUARDANT HEALTH EX.1009.481

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.

2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.

3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.

4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).

5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent CooperationTreaty.

6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).

7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.

8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.

9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112

BY EFS
Commissioner for Patents
Alexandria, VA

POWER OF ATTORNEY FROM ASSIGNEE
AND STATEMENT UNDER 37 C.F.R. § 3.73(b)

I hereby appoint pursuant to 37 C.F.R. § 3.73(b) the practitioners named below (and revoke any previous powers of attorney given) in connection with the subject application:

Name	Registration No.
John P. White	Reg. No. 28,678
Gary J. Gershik	Reg. No. 39,992
Norman H. Zivin	Reg. No. 25,385
Paul Teng	Reg. No. 40,837
Robert T. Maldonado	Reg. No. 38,232
Tonia A. Sayour	Reg. No. 58,404
Christopher C. Dunham	Reg. No. 22,031

as attorneys to represent the assignee identified below before the United States Patent and Trademark Office (USPTO) in connection with the subject application assigned to the assignee, as evidenced by the USPTO assignment records or assignment document(s) attached to this form in accordance with 37 C.F.R. § 3.73(b).

Please address all communications and direct all telephone calls regarding this application to:

GARY J. GERSHIK, ESQ.
COOPER & DUNHAM LLP
(CUSTOMER NUMBER 23432)
30 ROCKEFELLER PLAZA
NEW YORK, NEW YORK 10112
TEL. (212)278-0400

Cold Spring Harbor Laboratory, having a principal place of business at One Bungtown Road, Cold Spring Harbor, New York 11724, U.S.A., is the assignee of the entire right, title and interest in the subject application by virtue of an assignment, from James Hicks, Nicholas Navin, Jennifer Troge, Zihua Wang, and Michael Wigler to Cold Spring Harbor Laboratory of U.S. Serial No. 13/278,333, filed October 11, 2011, the parent of the subject application, which assignment was recorded with the U.S. Patent and Trademark Office in connection with the parent application on January 16, 2012 at Reel No. 027537, Frame 0950-0955. A copy of the recorded assignment is attached hereto as **Exhibit A**. The subject application is a continuation of U.S. Serial No. 13/278,333, and the attached assignment expressly refers to continuation applications. I am an official authorized to sign this Power Of Attorney on behalf of the assignee.

Date: 4/17/2019

Cold Spring Harbor Laboratory

By: _____

Vladimir Drozdoff, Ph.D., J.D.
Chief Patent Counsel
Office of Technology Transfer
Cold Spring Harbor Laboratory

PATENT ASSIGNMENT

Electronic Version v1.1
Stylesheet Version v1.1

SUBMISSION TYPE:	NEW ASSIGNMENT
NATURE OF CONVEYANCE:	ASSIGNMENT

CONVEYING PARTY DATA

Name	Execution Date
James Hicks	11/16/2011
Nicholas Navin	11/14/2011
Jennifer Troge	11/16/2011
Zihua Wang	11/16/2011
Michael Wigler	11/16/2011

RECEIVING PARTY DATA

Name:	Cold Spring Harbor Laboratory
Street Address:	One Bungtown Road
City:	Cold Spring Harbor
State/Country:	NEW YORK
Postal Code:	11724

PROPERTY NUMBERS Total: 1

Property Type	Number
Application Number:	13278333

CH $40.00 13278333

CORRESPONDENCE DATA

Fax Number: (212)391-0525
Phone: 2122780400
Email: patentdocketing@cooperdunham.com
Correspondence will be sent to the e-mail address first; if that is unsuccessful, it will be sent via US Mail.
Correspondent Name: Cooper & Dunham LLP
Address Line 1: 30 Rockefeller Plaza
Address Line 4: New York, NEW YORK 10112

U.S. Serial No.: 15/063,278
Applicant(s): James Hicks et al.
Filed: March 7, 2016
Exhibit A

ATTORNEY DOCKET NUMBER:	5981/81503-A/GJG/GC
NAME OF SUBMITTER:	Cindy Shu

Total Attachments: 4
source=120116_5981_81503_A_Assignment_GC_Page_1#page1.tif
source=120116_5981_81503_A_Assignment_GC_Page_2#page1.tif
source=120116_5981_81503_A_Assignment_GC_Page_3#page1.tif
source=120116_5981_81503_A_Assignment_GC_Page_4#page1.tif

Attorney Docket No. 5981/81503-A/GIG/ACK

Assignment of Non-Provisional Application

In consideration of One Dollar ($1.00), and other good and valuable consideration, the receipt of which is hereby acknowledged, we, the undersigned,

James Hicks residing at 3 Great Meadow Road, Locust Valley, NY 11560; Nicholas Navin residing at 1621 Wichita St., Houston, TX 77004; Jennifer Troge residing at 100 Simcoe St., Oyster Bay, NY 11771; Zihua Wang residing at 13 Meadowlawn St., Huntington, NY 11743; and Michael Wigler residing at 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Hereby sell, assign and transfer *to* Cold Spring Harbor Laboratory, *a corporation of the State of* New York, *having a place of business at* One Bungtown Road, Cold Spring Harbor, New York 11724 U.S.A, *in the County of* Suffolk *and State of* New York, *its successors, assigns and legal representatives, the entire right, title and interest for all countries, in and to any and all inventions which are disclosed and claimed, and any and all inventions which are disclosed but not claimed, in the application for United States Patent, which has been executed by the undersigned on* November 16, 2011; November 14, 2011; November 16, 2011; November 16, 2011; and November 16, 2011 *and is entitled*

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

(U.S. Serial No. 13/278,333, filed October 21, 2011, claiming the benefit of U.S. Provisional Application Nos. 61/510,579, filed July 22, 2011 and 61/406,067, filed October 22, 2010)

and in and to said application and all divisional, continuing, substitute, renewal, reissue, and all other applications for U.S. Letters Patent or other related property rights in any and all foreign countries which have been or shall be filed on any of said inventions disclosed in said application; and in and to all original and reissued patents or related foreign documents which have been or shall be issued on said inventions;

Authorize and request *the Commissioner for Patents of the United States to issue to said Assignee, the corporation above named, its successors, assigns and legal representatives, in accordance with this assignment, any and all United States Letters Patent on said inventions or any of them disclosed in said application;*

GUARDANT HEALTH EX1009.487
PATENT
REEL: 027607 FRAME: 0962

Agree *that said Assignee may apply for and receive foreign Letters Patent or rights of any other kind for said inventions, or any of them; and may claim, in applications for said foreign Letters Patent or other rights, the priority of the aforesaid United States patent application under the provisions of the International Convention of 1883 and later modifications thereof, under the Patent Cooperation Treaty, under the European Patent Convention or under any other available international agreement; and that, when requested, without charge to, but at the expense of, said Assignee, its successors, assigns and legal representatives, to carry out in good faith the intent and purpose of this assignment, the undersigned or the undersigned's executors or administrators will, for the United States and all foreign countries, execute all divisional, continuing, substitute, renewal, reissue, and all other patent applications or other documents on any and all said inventions; execute all rightful oaths, assignments, powers of attorney and other papers; communicate to said Assignee, its successors, assigns and representatives, all facts known and documents available to the undersigned relating to said inventions and the history thereof; testify in all legal proceedings; and generally do everything possible which said Assignee, its successors, assigns or representatives shall consider desirable for aiding in securing, maintaining and enforcing proper patent protection for said inventions and for vesting title to said inventions and all applications for patents or related foreign rights and all patents on said inventions, in said Assignee, its successors, assigns and legal representatives; and*

Covenant *with said Assignee, its successors, assigns and legal representatives that no assignment, grant, mortgage, license or other agreement affecting the rights and property herein conveyed has been made to others by the undersigned, and that full right to convey the same as herein expressed is possessed by the undersigned.*

Date: 11-16-11

Witness: _____ [signature]

Vladimir Drozdeff [printed name]

102 Cleveland Dr [address]

Centerport, NY 11721

_____ [L.S.]

James Hicks

JPW Rev. April 19, 2007

[LS]
Nicholas Navin

Date: 11-14-2011
Witness: _____ [signature]
Jill Waters [printed name]
1515 Holcombe Blvd [address]
Houston TX 77030

_____ [LS]
Jennifer Troge

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

_____ [LS]
Zihua Wang

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

_____ [LS]
Michael Wigler

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

_____ [LS]

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

PATENT
GUARDANT HEALTH EX.1009.489
REEL: 027537 FRAME: 0954

[LS]
Nicholas Navin

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

[LS]
Jennifer Troge

Date: Nov 16, 2011
Witness: _____ [signature]
Vladimir Drezdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

[LS]
Zihua Wang

Date: 11/16/2011
Witness: _____ [signature]
Vladimir Drezdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

[LS]
Michael Wigler

Date: 11/16/2011
Witness: _____ [signature]
Vladimir Drezdoff [printed name]
107 Cleveland Dr [address]
Centerport, NY 11721

[LS]

Date: _____
Witness: _____ [signature]
_____ [printed name]
_____ [address]
_____

GUARDANT HEALTH EX.1009.490
PATENT
RECORDED: 01/16/2012
REEL: 027537 FRAME: 0955

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension - 2 months with $0 paid	2252	1	300	300
Miscellaneous:				
STATUTORY OR TERMINAL DISCLAIMER	2814	1	160	160
		Total in USD ($)		**460**

Electronic Acknowledgement Receipt

EFS ID:	35758441
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	17-APR-2019
Filing Date:	07-MAR-2016
Time Stamp:	18:54:53
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$460
RAM confirmation Number	041819INTEFSW00005694033125
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.493

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	After Final Consideration Program Request	190417_81503-AA_AFCP_ADR.pdf	161288 228f5dfec0f589e3d0b1e21313f968a636c82523	no	2
Warnings:					
Information:					
2		190417_81503-AA_Amendment_ADR.pdf	1391218 dd22eadf52df081eac1c921b7f23fb67ab29a2f2	yes	23

Multipart Description/PDF files in .zip description

Document Description	Start	End
Response After Final Action	1	2
Drawings-other than black and white line drawings	3	3
Claims	4	11
Applicant Arguments/Remarks Made in an Amendment	12	20
Req to correct/update/change appl name until pay issue fee	21	22
Response After Final Action	23	23

Warnings:					
Information:					
3	Drawings-other than black and white line drawings	190417_81503-AA_Exhibit_1_ADR.pdf	1168657 7461ee57a166fcbcc278615597440921499d195a	no	8
Warnings:					
Information:					
4	Applicant Arguments/Remarks Made in an Amendment	190417_81503-AA_Exhibit_2_ADR.pdf	220789 59b2bc41a81c902239b27740ebbe751d00557132	no	27

GUARDANT HEALTH EX.1009.494

Warnings:					
Information:					
5	Terminal Disclaimer Filed	190417_81503-AA_Exhibit_3_ADR.pdf	1633515 284eadb4d1ea5bbec8014fcf6843aafa193657b6	no	3
Warnings:					
Information:					
6	Assignee showing of ownership per 37 CFR 3.73	190417_81503-AA_Exhibit_4_ADR.pdf	2246437 9efb15950b798296ac6d6995a70b64c7ee023398	no	3
Warnings:					
Information:					
7	Application Data Sheet	190417_81503-AA_Exhibit_5_ADR.pdf	3508285 2e7078ac9e30390028dc426131d34a01b2ee8daa	no	10
Warnings:					
Information:					
This is not an USPTO supplied ADS fillable form					
8	Power of Attorney	190417_81503-AA_Exhibit_6_ADR.pdf	2116649 bb6cb46980084157d72bfc2473789b2cd3f8d4de	no	2
Warnings:					
Information:					
9	Assignee showing of ownership per 37 CFR 3.73	190417_81503-AA_Exhibit_A_ADR.pdf	2329084 9705d00f70b5f5e4a1e0386e4ea640c1a027b215	no	6
Warnings:					
Information:					
10	Fee Worksheet (SB06)	fee-info.pdf	32679 5621724af011f838c2841a76d20c7dbc6c693653	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	14808601		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

DocCode – SCORE

SCORE Placeholder Sheet for IFW Content

Application Number: 15063278 Document Date: 04/17/2019

The presence of this form in the IFW record indicates that the following document type was received in electronic format on the date identified above. This content is stored in the SCORE database.

Since this was an electronic submission, there is no physical artifact folder, no artifact folder is recorded in PALM, and no paper documents or physical media exist. The TIFF images in the IFW record were created from the original documents that are stored in SCORE.

- Drawing

At the time of document entry (noted above):

- USPTO employees may access SCORE content via DAV or via the SCORE web page.
- External customers may access SCORE content via PAIR using the Supplemental Content tab.

Form Revision Date: March 1, 2019

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

POA ACCEPTANCE LETTER

OC000000107551137

Date Mailed: 04/19/2019

NOTICE OF ACCEPTANCE OF POWER OF ATTORNEY

This is in response to the Power of Attorney filed 04/17/2019.

The Power of Attorney in this application is accepted. Correspondence in this application will be mailed to the above address as provided by 37 CFR 1.33.

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at **(571) 272-4000** or **(571) 272-4200** or **1-888-786-0101**.

/tgebre/

GUARDANT HEALTH EX.1009.498

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

POWER OF ATTORNEY NOTICE

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

OC000000107551117

Date Mailed: 04/19/2019

NOTICE REGARDING CHANGE OF POWER OF ATTORNEY

This is in response to the Power of Attorney filed 04/17/2019.

- The Power of Attorney to you in this application has been revoked by the applicant. Future correspondence will be mailed to the new address of record(37 CFR 1.33).

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at **(571) 272-4000** or **(571) 272-4200** or **1-888-786-0101**.

/tgebre/

GUARDANT HEALTH EX.1009.499

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING or 371(c) DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	TOT CLAIMS	IND CLAIMS
15/063,278	03/07/2016	1634	730	81503-AA/GJG/AWG/AC	20	2

CONFIRMATION NO. 4038

CORRECTED FILING RECEIPT

23432
COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

OC000000107551185

Date Mailed: 04/19/2019

Receipt is acknowledged of this non-provisional patent application. The application will be taken up for examination in due course. Applicant will be notified as to the results of the examination. Any correspondence concerning the application must include the following identification information: the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. **If an error is noted on this Filing Receipt, please submit a written request for a Filing Receipt Correction. Please provide a copy of this Filing Receipt with the changes noted thereon. If you received a "Notice to File Missing Parts" for this application, please submit any corrections to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections**

Inventor(s)

James Hicks, Locust Valley, NY;
Nicholas Navin, Houston, TX;
Jennifer Troge, Oyster Bay, NY;
Zihua Wang, Huntington, NY;
Michael Wigler, Cold Spring Harbor, NY;

Applicant(s)

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY;

Assignment For Published Patent Application

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY

Power of Attorney: The patent practitioners associated with Customer Number 23432

Domestic Priority data as claimed by applicant

This application is a CON of 13/278,333 10/21/2011 PAT 9404156
which claims benefit of 61/510,579 07/22/2011
and claims benefit of 61/406,067 10/22/2010

Foreign Applications for which priority is claimed (You may be eligible to benefit from the **Patent Prosecution Highway** program at the USPTO. Please see http://www.uspto.gov for more information.) - None.
Foreign application information must be provided in an Application Data Sheet in order to constitute a claim to foreign priority. See 37 CFR 1.55 and 1.76.

Permission to Access Application via Priority Document Exchange: Yes

Permission to Access Search Results: Yes

GUARDANT HEALTH EX.1009.500

Applicant may provide or rescind an authorization for access using Form PTO/SB/39 or Form PTO/SB/69 as appropriate.

If Required, Foreign Filing License Granted: 03/22/2016
The country code and number of your priority application, to be used for filing abroad under the Paris Convention, is **US 15/063,278**
Projected Publication Date: Not Applicable
Non-Publication Request: No
Early Publication Request: No
**** SMALL ENTITY ****
Title

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Preliminary Class

435

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications: No

PROTECTING YOUR INVENTION OUTSIDE THE UNITED STATES

Since the rights granted by a U.S. patent extend only throughout the territory of the United States and have no effect in a foreign country, an inventor who wishes patent protection in another country must apply for a patent in a specific country or in regional patent offices. Applicants may wish to consider the filing of an international application under the Patent Cooperation Treaty (PCT). An international (PCT) application generally has the same effect as a regular national patent application in each PCT-member country. The PCT process **simplifies** the filing of patent applications on the same invention in member countries, but **does not result** in a grant of "an international patent" and does not eliminate the need of applicants to file additional documents and fees in countries where patent protection is desired.

Almost every country has its own patent law, and a person desiring a patent in a particular country must make an application for patent in that country in accordance with its particular laws. Since the laws of many countries differ in various respects from the patent law of the United States, applicants are advised to seek guidance from specific foreign countries to ensure that patent rights are not lost prematurely.

Applicants also are advised that in the case of inventions made in the United States, the Director of the USPTO must issue a license before applicants can apply for a patent in a foreign country. The filing of a U.S. patent application serves as a request for a foreign filing license. The application's filing receipt contains further information and guidance as to the status of applicant's license for foreign filing.

Applicants may wish to consult the USPTO booklet, "General Information Concerning Patents" (specifically, the section entitled "Treaties and Foreign Patents") for more information on timeframes and deadlines for filing foreign patent applications. The guide is available either by contacting the USPTO Contact Center at 800-786-9199, or it can be viewed on the USPTO website at http://www.uspto.gov/web/offices/pac/doc/general/index.html.

For information on preventing theft of your intellectual property (patents, trademarks and copyrights), you may wish to consult the U.S. Government website, http://www.stopfakes.gov. Part of a Department of Commerce initiative, this website includes self-help "toolkits" giving innovators guidance on how to protect intellectual property in specific

GUARDANT HEALTH EX.1009.501

countries such as China, Korea and Mexico. For questions regarding patent enforcement issues, applicants may call the U.S. Government hotline at 1-866-999-HALT (1-866-999-4258).

LICENSE FOR FOREIGN FILING UNDER

Title 35, United States Code, Section 184

Title 37, Code of Federal Regulations, 5.11 & 5.15

GRANTED

The applicant has been granted a license under 35 U.S.C. 184, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" followed by a date appears on this form. Such licenses are issued in all applications where the conditions for issuance of a license have been met, regardless of whether or not a license may be required as set forth in 37 CFR 5.15. The scope and limitations of this license are set forth in 37 CFR 5.15(a) unless an earlier license has been issued under 37 CFR 5.15(b). The license is subject to revocation upon written notification. The date indicated is the effective date of the license, unless an earlier license of similar scope has been granted under 37 CFR 5.13 or 5.14.

This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

The grant of a license does not in any way lessen the responsibility of a licensee for the security of the subject matter as imposed by any Government contract or the provisions of existing laws relating to espionage and the national security or the export of technical data. Licensees should apprise themselves of current regulations especially with respect to certain countries, of other agencies, particularly the Office of Defense Trade Controls, Department of State (with respect to Arms, Munitions and Implements of War (22 CFR 121-128)); the Bureau of Industry and Security, Department of Commerce (15 CFR parts 730-774); the Office of Foreign AssetsControl, Department of Treasury (31 CFR Parts 500+) and the Department of Energy.

NOT GRANTED

No license under 35 U.S.C. 184 has been granted at this time, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" DOES NOT appear on this form. Applicant may still petition for a license under 37 CFR 5.12, if a license is desired before the expiration of 6 months from the filing date of the application. If 6 months has lapsed from the filing date of this application and the licensee has not received any indication of a secrecy order under 35 U.S.C. 181, the licensee may foreign file the application pursuant to 37 CFR 5.15(b).

SelectUSA

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The U.S. offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to promote and facilitate business investment. SelectUSA provides information assistance to the international investor community; serves as an ombudsman for existing and potential investors; advocates on behalf of U.S. cities, states, and regions competing for global investment; and counsels U.S. economic development organizations on investment attraction best practices. To learn more about why the United States is the best country in the world to develop

GUARDANT HEALTH EX.1009.502

technology, manufacture products, deliver services, and grow your business, visit http://www.SelectUSA.gov or call +1-202-482-6800.

GUARDANT HEALTH EX.1009.503

Application Number * 15/063,278 *	Application/Control No. 15/063,278	Applicant(s)/Patent under Reexamination Hicks et al.	
	Examiner LU, FRANK WEI MIN	Art Unit 1634	

Document Code - DISQ	**Internal Document - DO NOT MAIL**

TERMINAL DISCLAIMER	☑ APPROVED	☐ DISAPPROVED
Date Filed: 17 April 2019	**This patent is subject to a Terminal Disclaimer**	

Approved/Disapproved by:

/FELICIA D ROBERTS/

Technology Center: OPLC

Telephone: (571)272-0550

9,404,156

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 04/25/2019

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
04/25/2019	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.505

DETAILED ACTION

Advisory Action

1. The proposed amendment filed on April 19, 2019 has been fully considered but will not be entered because they raise new issues that would require further consideration and/or search.

Response to Arguments

In page 15, third paragraph bridging to page 20, fourth paragraph of applicant's remarks, applicant argues that the proposed amendment has overcome all objections and rejections under 35 U.S.C 101 and 112 (b) and the double patenting rejection.

The argument has been fully considered but it is not persuasive toward the withdrawal of the objections and rejections because applicant's argument with respect to claims 68-75 and 77-87 are moot since applicant has amended claims 68, 69, 75, 77, 81, and 85. Claims 68, 69, 75, 77, 81, and 85 in the proposed amendment raise new issues that would require further consideration and/or search. For example, the phrase "wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome" in claim 68 or 75 of the proposed amendment is a new limitation and has not been found in the amendment filed on October 17, 2018, which is the amendment used for the final rejection. The double patenting rejection will be withdrawn if the terminal disclaimer has been approved by the office.

2. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/FRANK W LU/
Primary Examiner, Art Unit 1634
April 24, 2019

Advisory Action Before the Filing of an Appeal Brief	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

--The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

THE REPLY FILED 19 April 2019 FAILS TO PLACE THIS APPLICATION IN CONDITION FOR ALLOWANCE.

NO NOTICE OF APPEAL FILED

1. ☑ The reply was filed after a final rejection. No Notice of Appeal has been filed. To avoid abandonment of this application, applicant must timely file one of the following replies: (1) an amendment, affidavit, or other evidence, which places the application in condition for allowance; (2) a Notice of Appeal (with appeal fee) in compliance with 37 CFR 41.31; or (3) a Request for Continued Examination (RCE) in compliance with 37 CFR 1.114 if this is a utility or plant application. Note that RCEs are not permitted in design applications. The reply must be filed within one of the following time periods:
   a) ☑ The period for reply expires 5 months from the mailing date of the final rejection.
   b) ☐ The period for reply expires on: (1) the mailing date of this Advisory Action; or (2) the date set forth in the final rejection, whichever is later. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of the final rejection.
   c) ☐ A prior Advisory Action was mailed more than 3 months after the mailing date of the final rejection in response to a first after-final reply filed within 2 months of the mailing date of the final rejection.The current period for reply expires ____ months from the mailing date of *the prior Advisory Action* or SIX MONTHS from the mailing date of the final rejection, whichever is earlier.
   Examiner Note: If box 1 is checked, check either box (a), (b) or (c). ONLY CHECK BOX (b) WHEN THIS ADVISORY ACTION IS THE FIRST RESPONSE TO APPLICANTS FIRST AFTER-FINAL REPLY WHICH WAS FILED WITHIN TWO MONTHS OF THE FINAL REJECTION. ONLY CHECK BOX (c) IN THE LIMITED SITUATION SET FORTH UNDER BOX (c). See MPEP 706.07(f).

Extensions of time may be obtained under 37 CFR 1.136(a). The date on which the petition under 37 CFR 1.136(a) and the appropriate extension fee have been filed is the date for purposes of determining the period of extension and the corresponding amount of the fee. The appropriate extension fee under 37 CFR 1.17(a) is calculated from: (1) the expiration date of the shortened statutory period for reply originally set in the final Office action; or (2) as set forth in (b) or (c) above, if checked. Any reply received by the Office later than three months after the mailing date of the final rejection, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

NOTICE OF APPEAL

2. ☐ The Notice of Appeal was filed on _____. A brief in compliance with 37 CFR 41.37 must be filed within two months of the date of filing the Notice of Appeal (37 CFR 41.37(a)), or any extension thereof (37 CFR 41.37(e)), to avoid dismissal of the appeal. Since a Notice of Appeal has been filed, any reply must be filed within the time period set forth in 37CFR 41.37(a).

AMENDMENTS

3. ☑ The proposed amendments filed after a final rejection, but prior to the date of filing a brief, will not be entered because
   a) ☑ They raise new issues that would require further consideration and/or search (see NOTE below);
   b) ☐ They raise the issue of new matter (see NOTE below);
   c) ☐ They are not deemed to place the application in better form for appeal by materially reducing or simplifying the issues for appeal; and/or
   d) ☐ They present additional claims without canceling a corresponding number of finally rejected claims.
   NOTE: See attached office action (See 37CFR 1.116 and 41.33(a)).
4. ☐ The amendments are not in compliance with 37CFR 1.121. See attached Notice of Non-Compliant Amendment (PTOL-324).
5. ☐ Applicants reply has overcome the following rejection(s): _____
6. ☐ Newly proposed or amended claim(s) _____ would be allowable if submitted in a separate, timely filed amendment canceling the non-allowable claim(s).
7. ☑ For purposes of appeal, the proposed amendment(s):(a) ☑ will not be entered, or (b) ☐ will be entered, and an explanation of how the new or amended claims would be rejected is provided below or appended.

AFFIDAVIT OR OTHER EVIDENCE

8. ☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____
9. ☐ The affidavit or other evidence filed after final action, but before or on the date of filing a Notice of Appeal will not be entered because applicant failed to provide a showing of good and sufficient reasons why the affidavit or other evidence is necessary and was not earlier presented. See 37 CFR 1.116(e).
10. ☐ The affidavit or other evidence filed after the date of filing the Notice of Appeal, but prior to the date of filing a brief, will not be entered because the affidavit or other evidence failed to overcome all rejections under appeal and/or appellant fails to provide a showing of good and sufficient reasons why it is necessary and was not earlier presented. See 37 CFR 41.33(d)(1).
11. ☐ The affidavit or other evidence is entered. An explanation of the status of the claims after entry is below or attached.

REQUEST FOR RECONSIDERATION/OTHER

12. ☐ The request for reconsideration has been considered but does NOT place the application in condition for allowance because: _____.
13. ☐ Note the attached Information *Disclosure Statement*(s). (PTO/SB/08) Paper No(s). _____
14. ☑ Other: PTOL-2323 and PTOL-413B.

STATUS OF CLAIMS

15. The status of the claim(s) is (or will be) as follows:
    Claim(s) allowed:_____.
    Claim(s) objected to:_____.
    Claim(s) rejected:68-75 and 77-87.
    Claim(s) withdrawn from consideration:76.

/FRANK W LU/ Primary Examiner, Art Unit 1634	April 24, 2019

GUARDANT HEALTH EX.1009.508

Examiner-Initiated Interview Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

All participants (applicant, applicant's representative, PTO personnel):

(1) FRANK LU. (3) _____.

(2) Mr. Gary Gershik (Reg. No. 39,992). (4) _____.

Date of Interview: 23 April 2019.

Type: ☑ Telephonic ☐ Video Conference
☐ Personal [copy given to: ☐ applicant ☐ applicant's representative]

Exhibit shown or demonstration conducted: ☐ Yes ☐ No.
If Yes, brief description: _____.

Issues Discussed ☐ 101 ☐ 112 ☐ 102 ☐ 103 ☑ Others
(For each of the checked box(es) above, please describe below the issue and detailed description of the discussion)

Claim(s) discussed: 68-75 and 77-85.

Identification of prior art discussed: _____.

Substance of Interview
(For each issue discussed, provide a detailed description and indicate if agreement was reached. Some topics may include: identification or clarification of a reference or a portion thereof, claim interpretation, proposed amendments, arguments of any applied references etc...)

See Continuation Sheet.

Applicant recordation instructions: It is not necessary for applicant to provide a separate record of the substance of interview.

Examiner recordation instructions: Examiners must summarize the substance of any interview of record. A complete and proper recordation of the substance of an interview should include the items listed in MPEP 713.04 for complete and proper recordation including the identification of the general thrust of each argument or issue discussed, a general indication of any other pertinent matters discussed regarding patentability and the general results or outcome of the interview, to include an indication as to whether or not agreement was reached on the issues raised.

☐ Attachment

/FRANK W LU/ Primary Examiner, Art Unit 1634	April 23, 2019

GUARDANT HEALTH EX.1009.509

Continuation of Substance of Interview including description of the general nature of what was agreed to if an agreement was reached, or any other comments: The examiner called Mr. Gershik. Since applicant has amended claims 68, 69, 75, 77, 81, and 85, claims 68, 69, 75, 77, 81, and 85 in the proposed amendment raise new issues that would require further consideration and/or search. For example, the phrase “wherein more than one count are counts, and each count represents an estimate of the genomic copy number at a location of the genome” in claim 68 or 75 of the proposed amendment is a new limitation and has not been found in the amendment filed on October 17, 2018, which is the amendment used for the final rejection. The double patenting rejection will be withdrawn if the terminal disclaimer has been approved by the office.

AFCP 2.0 Decision	Application No.	Applicant(s)	
	15/063,278	Hicks et al.	
	Examiner	Art Unit	AIA (FITF) Status
	FRANK W LU	1634	No

This is in response to the After Final Consideration Pilot request filed 19 April 2019.

1. **Improper Request** – The AFCP 2.0 request is improper for the following reason(s) and the after final amendment submitted with the request will be treated under pre-pilot procedure.

   ☐ An AFCP 2.0 request form PTO/SB/434 (or equivalent document) was not submitted.

   ☐ A non-broadening amendment to at least one independent claim was not submitted.

   ☐ A proper AFCP 2.0 request was submitted in response to the most recent final rejection.

   ☐ Other: _____

2. **Proper Request**

   A. After final amendment submitted with the request will not be treated under AFCP 2.0.
   The after final amendment cannot be reviewed and a search conducted within the guidelines of the pilot program.

   ☐ The after final amendment will be treated under pre-pilot procedure.

   B. Updated search and/or completed additional consideration.
   The examiner performed an updated search and/or completed additional consideration of the after final amendment within the time authorized for the pilot program. The result(s) of the updated search and/or completed additional consideration are:

   ☐ 1. All of the rejections in the most recent final Office action are overcome and a Notice of Allowance is issued herewith.

   ☐ 2. The after final amendment would not overcome all of the rejections in the most recent final Office action . See attached interview summary for further details.

   ☑ 3. The after final amendment was reviewed, and it raises a new issue(s). See attached interview summary for further details.

   ☐ 4. The after final amendment raises new issues, but would overcome all of the rejections in the most recent final Office action. A decision on determining allowability could not be made within the guidelines of the pilot. See attached interview summary for further details, including any newly discovered prior art.

   ☐ 5. Other: _____

   Examiner Note: Please attach an interview summary when necessary as described above.

GUARDANT HEALTH EX.1009.511

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
April 17, 2019

BY EFS
Commissioner for Patents
Alexandria, VA

DO NOT ENTER: /F.W.L/

AMENDMENT IN RESPONSE TO NOVEMBER 30, 2018 FINAL OFFICE ACTION AS A REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0 AND PETITION FOR A TWO-MONTH EXTENSION OF TIME

This Amendment is in response to the November 30, 2018 Final Office Action issued by the United States Patent and Trademark Office in connection with the above-identified application. A response to the November 30, 2018 Final Office Action was originally due February 28, 2019. Applicants hereby petition for a two-month extension of time. The required fee for a two-month extension of time is THREE HUNDRED DOLLARS ($300.00) for a small entity and authorization is hereby given to charge this fee to Deposit Account No. 03-3125. With a two-month extension of time, a response to the November 30, 2018 Final Office Action is now due April 30, 2019. Accordingly, this Amendment is being timely filed.

Amendments to the Drawings are on page **3** of this paper.

Amendments to the Claims begin on page **4** of this paper.

Remarks begin on page **12** of this paper.

A **Terminal Disclaimer** is referred to on page **19** of this paper and is attached hereto as **Exhibit 3.**

A **Request to Update the Name of the Applicant** begins on page **21** of this paper.

PTO/SB/30 (07-09)
Approved for use through 07/31/2012. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Request for Continued Examination (RCE) Transmittal

Address to:
Mail Stop RCE
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Application Number	15/063,278
Filing Date	March 7, 2016
First Named Inventor	James Hicks
Art Unit	1634
Examiner Name	Frank W. M. Lu
Attorney Docket Number	81503-AA/GJG/ADR

This is a Request for Continued Examination (RCE) under 37 CFR 1.114 of the above-identified application.
Request for Continued Examination (RCE) practice under 37 CFR 1.114 does not apply to any utility or plant application filed prior to June 8, 1995, or to any design application. See Instruction Sheet for RCEs (not to be submitted to the USPTO) on page 2.

1. Submission required under 37 CFR 1.114 Note: If the RCE is proper, any previously filed unentered amendments and amendments enclosed with the RCE will be entered in the order in which they were filed unless applicant instructs otherwise. If applicant does not wish to have any previously filed unentered amendment(s) entered, applicant must request non-entry of such amendment(s).

   a. [x] Previously submitted. If a final Office action is outstanding, any amendments filed after the final Office action may be considered as a submission even if this box is not checked.

      i. [ ] Consider the arguments in the Appeal Brief or Reply Brief previously filed on _____

      ii. [x] Other April 17, 2019 Amendment in Response to November 30, 2018 Final Office Action

   b. [ ] Enclosed

      i. [ ] Amendment/Reply

      ii. [ ] Affidavit(s)/ Declaration(s)

      iii. [ ] Information Disclosure Statement (IDS)

      iv. [ ] Other _____

2. Miscellaneous

   a. [ ] Suspension of action on the above-identified application is requested under 37 CFR 1.103(c) for a period of _____ months. (Period of suspension shall not exceed 3 months; Fee under 37 CFR 1.17(i) required)

   b. [ ] Other _____

3. Fees The RCE fee under 37 CFR 1.17(e) is required by 37 CFR 1.114 when the RCE is filed.

   a. [x] The Director is hereby authorized to charge the following fees, any underpayment of fees, or credit any overpayments, to Deposit Account No. 03-3125.

      i. [x] RCE fee required under 37 CFR 1.17(e)

      ii. [x] Extension of time fee (37 CFR 1.136 and 1.17)

      iii. [ ] Other _____

   b. [ ] Check in the amount of $ _____ enclosed

   c. [ ] Payment by credit card (Form PTO-2038 enclosed)

WARNING: Information on this form may become public. Credit card information should not be included on this form. Provide credit card information and authorization on PTO-2038.

SIGNATURE OF APPLICANT, ATTORNEY, OR AGENT REQUIRED

Signature	/Gary J. Gershik/	Date	May 30, 2019
Name (Print/Type)	Gary J. Gershik	Registration No.	39992

CERTIFICATE OF MAILING OR TRANSMISSION

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as first class mail in an envelope addressed to: Mail Stop RCE, Commissioner for Patents, P. O. Box 1450, Alexandria, VA 22313-1450 or facsimile transmitted to the U.S. Patent and Trademark Office on the date shown below.

Signature			
Name (Print/Type)		Date	

This collection of information is required by 37 CFR 1.114. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.11 and 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Mail Stop RCE, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

Instruction Sheet for RCEs

(not to be submitted to the USPTO)

NOTES:

An RCE is not a new application, and filing an RCE will not result in an application being accorded a new filing date.

Filing Qualifications:

The application must be a utility or plant application filed on or after June 8, 1995. The application cannot be a provisional application, a utility or plant application filed before June 8, 1995, a design application, or a patent under reexamination. See 37 CFR 1.114(e).

Filing Requirements:

Prosecution in the application must be closed. Prosecution is closed if the application is under appeal, or the last Office action is a final action, a notice of allowance, or an action that otherwise closes prosecution in the application (e.g., an Office action under *Ex parte Quayle*). See 37 CFR 1.114(b).

A submission and a fee are required at the time the RCE is filed. If reply to an Office action under 35 U.S.C. 132 is outstanding (e.g., the application is under final rejection), the submission must meet the reply requirements of 37 CFR 1.111. If there is no outstanding Office action, the submission can be an information disclosure statement, an amendment, new arguments, or new evidence. See 37 CFR 1.114(c). The submission may be a previously filed amendment (e.g., an amendment after final rejection).

WARNINGS:

Request for Suspension of Action:

All RCE filing requirements must be met before suspension of action is granted. A request for a suspension of action under 37 CFR 1.103(c) does not satisfy the submission requirement and does not permit the filing of the required submission to be suspended.

Improper RCE will NOT toll Any Time Period:

Before Appeal - If the RCE is improper (e.g., prosecution in the application is not closed or the submission or fee has not been filed) and the application is not under appeal, the time period set forth in the last Office action will continue to run and the application will be abandoned after the statutory time period has expired if a reply to the Office action is not timely filed. No additional time will be given to correct the improper RCE.

Under Appeal - If the RCE is improper (e.g., the submission or the fee has not been filed) and the application is under appeal, the improper RCE is effective to withdraw the appeal. Withdrawal of the appeal results in the allowance or abandonment of the application depending on the status of the claims. If there are no allowed claims, the application is abandoned. If there is at least one allowed claim, the application will be passed to issue on the allowed claim(s). See MPEP 1215.01.

See MPEP 706.07(h) for further information on the RCE practice.

GUARDANT HEALTH EX.1009.515

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension - 3 months with $300 paid	2253	1	400	400
Miscellaneous:				
RCE- 1ST REQUEST	2801	1	650	650
		Total in USD ($)		**1050**

Electronic Acknowledgement Receipt

EFS ID:	36161181
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	30-MAY-2019
Filing Date:	07-MAR-2016
Time Stamp:	18:34:16
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$1050
RAM confirmation Number	053119INTEFSW00005185033125
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.519

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Extension of Time	190530_81503-AA_Extension_of_Time_ADR.pdf	71441 4b5e19c781ce39b5055ea0a347a90500dd16f04c	no	2
Warnings:					
Information:					
2	Request for Continued Examination (RCE)	190530_81503-AA_RCE_ADR2.pdf	1215229 fd70a203f4e56aa5e03eac78a5fb3f593ae250e1	no	3
Warnings:					
This is not a USPTO supplied RCE SB30 form.					
Information:					
3	Fee Worksheet (SB06)	fee-info.pdf	32579 11a155c5c151fd3f879c4ade127e0c1aeb36775c	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	1319249		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
May 30, 2019

BY EFS
Commissioner for Patents
Alexandria, VA

PETITION FOR EXTENSION OF TIME

A Response to the outstanding November 30, 2018 Final Office Action was originally due February 28, 2019. In the April 17, 2019 Amendment in Response to November 30, 2018 Final Office Action, applicants petitioned for a two-month extension of time and paid the required fee. Applicants hereby petition for a third month extension of time. The fee for a third month extension of time is FOUR HUNDRED DOLLARS ($400.00) for a small entity ($700.00 minus $300.00) and authorization is hereby given to charge this amount to Deposit Account No. 03-3125. With a third month extension of time, the deadline for taking further action is now November 30, 2018 Final Office Action is now due May 30, 2019. Accordingly, the subject application is pending today, May 29, 2019, for the purpose of filing the accompanying Request for Continued Examination.

No fee, other than the $400.00 fee for the filing of a third-month extension of time for which the authorization to charge Deposit Account No. 03-3125 has been given on page 1 of this Petition, is deemed necessary in connection with the filing of this Petition. If any fee is required, authorization is hereby given to charge the amount of any such required fee to Deposit Account No. 03-3125.

Respectfully submitted,

Gary J. Gershik

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

2019-05-30

Aditya Ramanathan Date

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 10/03/2019

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

MAIL DATE	DELIVERY MODE
10/03/2019	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

GUARDANT HEALTH EX.1009.523

Office Action Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☑ Responsive to communication(s) filed on 5/30/2019.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☐ This action is **FINAL.** 2b) ☑ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5) ☑ Claim(s) 68-87 is/are pending in the application.

5a) Of the above claim(s) 76 is/are withdrawn from consideration.

6) ☐ Claim(s) _____ is/are allowed.

7) ☑ Claim(s) 68-75 and 77-87 is/are rejected.

8) ☐ Claim(s) _____ is/are objected to.

9) ☐ Claim(s) _____ are subject to restriction and/or election requirement

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to **PPHfeedback@uspto.gov.**

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☑ The drawing(s) filed on 4/17/2019 is/are: a)☑ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)

2) ☑ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date 2/25/2019.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date _____.

4) ☐ Other: _____.

GUARDANT HEALTH EX.1009.524

DETAILED ACTION

CONTINUED EXAMINATION UNDER 37 CFR 1.114 AFTER FINAL REJECTION

1. A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission of RCE filed on April 17, 2019 and the amendment filed on May 30, 2019 have been entered. The claims pending in this application are claims 68-87 wherein claim 76 has been withdrawn due to the restriction requirement mailed on January 18, 2018. Rejection and/or objection not reiterated from the previous office action are hereby withdrawn in view of applicant's amendment filed on April 17, 2019. Claims 68-75 and 77-87 will be examined.

Drawings

2. New Figures 7A to 7C, 8, 9A, 9B, 10A and 10B submitted on April 17, 2019 have been accepted by the office.

Claim Rejections - 35 USC § 112

3. The following is a quotation of 35 U.S.C. 112(b):

> (b) CONCLUSION.—The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the inventor or a joint inventor regards as the invention.

The following is a quotation of 35 U.S.C. 112 (pre-AIA), second paragraph:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

4. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 112(b) or 35 U.S.C. 112 (pre-AIA), second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which the inventor or a joint inventor, or for pre-AIA the applicant regards as the invention.

5. Claim 68 is rejected as vague and indefinite. Since step f) only requires counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the genome, a count for the same location on the genome can be obtained. Therefore, a count for each location on the genome and genomic copy number information from the genomic nucleic acids (eg., different genomic nucleic acids) cannot be obtained only based on step f). Furthermore, it is unclear that more than one count is obtained from where and what is the relationship between the more than one count and each count. Please clarify.

6. Claim 75 is rejected as vague and indefinite because it is unclear how mRNA copy number information can obtained from the mRNA transcripts only based on step e) because the claim does not indicate what is the relationship between mRNA copy number information and the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library. Since step e) only requires counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, only a count for the same location on the cDNA library can be obtained.Thus, it is unclear that more than one count are from where, and what is the relationship between the more than one count and each count. Please clarify.

Claim Rejections - 35 USC § 101

7. 35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

8. Claims 68-75 and 77-87 are rejected under 35 U.S.C. 101 because the claimed invention is directed to a judicial exception (i.e., a law of nature, a natural phenomenon, or an abstract idea) without significantly more.

The processes of claims 68-74 and 77-87 are directed to a process of obtaining genomic copy number information from genomic nucleic acids wherein steps e) and f) in independent claim 68 and dependent claims 69-72, 74, and 82 fall within the mental process grouping of abstract ideas. The process of claim 75 is directed to a process of obtaining mRNA copy number information from mRNA transcripts wherein steps d) and e) in independent claim 75 fall within the mental process grouping of abstract ideas. **Note that steps in a claim that recite the manipulation of information through a series of mental steps would be considered a single abstract idea for purposes of analysis** (see MPEP 2106.04).

This judicial exception, the abstract idea, is not integrated into a practical application because the additional elements, steps e) and f) in independent claim 68, dependent claims 69-72, 74, and 82 and steps d) and e) in independent claim 75 are mental steps that do not integrate the recited judicial exception into a practical application.

The claims do not include additional elements that are sufficient to amount to significantly more than the judicial exception because independent claim 68, the steps a) to d) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see claims 1-21 of US Patent No. 9,670,536 B2 with a priority date of September 21, 2010) and pages 119 and 120, online methods and Figure 1 from Hiatt *et*

al., Nature Methods, 7, 119-122, published online on January 17, 2010) while, in independent claim 75, the steps a) to c) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see Figures 1a, 1c and 1d in page 529 and online methods from Plessy *et al.,* Nature Methods, 7, 528-534, published online on June 13, 2010). The conventional steps do not comprise an inventive concept that transforms the claimed abstract ideas into a patent-eligible application of the abstract ideas. The claimed subject matters of claims 68-75 and 77-87 are processes directed to abstract ideas. When recited at this high level of generality, there is no meaningful limitation, such as a particular or unconventional machine or a transformation of a particular article, in this step that distinguishes it from well-understood, routine, and conventional data gathering activity engaged in by scientists prior to applicant's invention, and at the time the application was filed, *e.g.*, the routine and conventional techniques for performing steps a) to d) of claim 68 or steps a) to c) of claim 75. Further, it is well established that the mere physical or tangible nature of additional elements such as steps a) to d) of claim 68 or steps a) to c) of claim 75 do not automatically confer eligibility on a claim directed to an abstract idea (*see, e.g.*, *Alice Corp. v. CLS Bank Int'l,* 134 S. Ct. 2347, 2358-59 (2014)). Therefore, the claimed subject matters of claims 68-75 and 77-87 are without limitations that indicate that the claims are no different from the process claims directed to abstract ideas and are patent-ineligible.

Response to Arguments

In page 17, third paragraph bridging to page 17, second paragraph of applicant' remarks, applicant argues that, in view of "2019 Revised Patent Subject Matter Eligibility Guidance" on January 7, 2019, "the pending claims are directed to a method of sequencing that (i) comprises specific steps (specific elements) (a)-(c) that transform genomic nucleic acids to generate new,

non-naturally occurring molecules (unique tagged nucleic acid molecules) that are sequenced and mapped to a location on the genome, and then (ii) a step to count the different, mapped, transformed molecules for the practical purpose (application) of obtaining genomic copy number information, for example, to generate a genomic profile of a cell (for example a cancer cell) or of a population of cells. In this way, the specific transformation steps (elements) required by the claimed method integrate the mapping and counting steps into a practical application, in a manner that avoids distortion of copy number by amplification of the genomic nucleic acids. Accordingly, in light of the Revised Guidance, applicants submit that the pending claims comply with 35 U.S.C. § 101 and respectfully request that this rejection be withdrawn".

The above argument has been fully considered but it is not persuasive toward the withdrawal of the rejection because in view of 2019 Revised Patent Subject Matter Eligibility Guidance mentioned by applicant, steps e) and f) in independent claim 68, dependent claims 69-72, 74, and 82 and steps d) and e) in independent claim 75 are mental steps that do not integrate the recited judicial exception into a practical application (for detail, see above rejection).

Conclusion

9. No claim is allowed.

10. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993)(See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/FRANK W LU/
Primary Examiner, Art Unit 1634
September 30, 2019

Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	Art Unit
	FRANK W LU	1634

CPC - Searched*		
Symbol	Date	Examiner
Update	09/30/2019	/FL/

CPC Combination Sets - Searched*		
Symbol	Date	Examiner
Update	09/30/2019	/FL/

US Classification - Searched*			
Class	Subclass	Date	Examiner
Update		09/30/2019	/FL/

* See search history printout included with this form or the SEARCH NOTES box below to determine the scope of the search.

Search Notes		
Search Notes	Date	Examiner
Update	09/30/2019	/FL/

Interference Search			
US Class/CPC Symbol	US Subclass/CPC Group	Date	Examiner

/FRANK LU/ Primary Examiner, Art Unit 1634	September 30, 2019

GUARDANT HEALTH EX.1009.531

Substitute Form PTO-1449 (Modified) U.S. Department of Commerce Patent and Trademark Office	Attorney Docket No. 81503-AA/GJG/ADR	Application No. 15/063,278
Information Disclosure Statement by Applicant (Use several sheets if necessary)	First Named Inventor James HICKS	Confirmation Number 4038
(37 CFR §1.98(b))	Filing Date March 7, 2016	Group Art Unit 1634

U.S. Patent Documents				
Examiner Initial	Cite ID[1]	Document Number Number-Kind Code[2] (if known)	Publication Date MM/DD/YYYY	Inventor(s) or Applicant(s)
	3.	2008-0261204	10/23/2008	LEXOW
	4.	2007-0020640	01/25/2007	MCCLOSKEY et al.

FOREIGN PATENT DOCUMENTS						
Examiner Initial	Cite ID[1]	Foreign Patent Document Country Code[3] Number[4] Kind Code[5] (if known)	Publication Date MM/DD/YYYY	Inventor(s)	Country or Patent Office[3]	T[6]
	5.	DE 10 2008 025 656 A1	12/03/2009	Bjorn ROTTER	DE	

NON PATENT LITERATURE DOCUMENTS		
Examiner Initial	Cite ID[1]	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc.) date, page(s), volume-issue number(s), publisher, city and/or country where published.
	1.	Communication of a notice of opposition, issued by the European Patent Office on February 1, 2019, concerning European Patent No. EP2630263
	2.	Notice of opposition to a European Patent, filed January 25, 2019, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263
	6.	Chiang et al. (2008) "High-resolution Mapping of Copy-number Alterations with Massively Parallel Sequencing", Nature Methods 6(1):99-103
	7.	Hug and Schuler (2003) "Measurement of the Number of Molecules of a Single mRNA Species in a Complex mRNA Preparation", J. theor. Biol. 221:615-624
	8.	Meyerson et al. (2010) "Advances in Understanding Cancer Genomes Through Second-Generation Sequencing", Nature Reviews 11:685-696

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

Examiner Signature /FRANK W LU/	Date Considered 09/30/2019

***EXAMINER:** Initial if citation considered, whether or not citation is in conformance with MPEP 609: Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant. [1] Applicant's unique citation designation number (optional). [2] See Kinds of Codes of USPTO Patent Documents at www.uspto.gov or MPEP 901.04. [3] Enter Office that issued the document, by the two-letter code (WIPO Standard ST.3). [4] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [5] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [6] Applicant is to place a check mark here if English Language Translation is attached.

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☑ SMALL ☐ MICRO

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A		N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A		N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*		x $40 =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*		x $210 =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).				
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))					
* If the difference in column 1 is less than zero, enter "0" in column 2.				TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT	04/03/2020	CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0		x $50 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	*** 3	= 0		x $230 =	0
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	0

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT		CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=		x $0 =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=		x $0 =	
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.	LIE
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".	/CHRISTINE V MOORE/
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".	
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.	

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
April 3, 2020

BY EFS
Commissioner for Patents
Alexandria, VA

AMENDMENT IN RESPONSE TO OCTOBER 3, 2019 NON-FINAL OFFICE ACTION, PETITION FOR A THREE-MONTH EXTENSION OF TIME, SUMMARY OF FEBRUARY 5, 2020 EXAMINER INTERVIEW, AND SUMMARY OF APRIL 3, 2020 EXAMINER INTERVIEW

This Amendment is in response to the October 3, 2019 Office Action issued by the United States Patent and Trademark Office in connection with the above-identified application. A response to the October 3, 2019 Office Action was originally due January 3, 2020. Applicants hereby petition for a three-month extension of time. The required fee for a two-month extension of time is SEVEN HUNDRED DOLLARS ($700.00) for a small entity and authorization is hereby given to charge this fee to Deposit Account No. 03-3125. With a three-month extension of time, a response to the October 3, 2019 Office Action is now due April 3, 2020. Accordingly, this Amendment is being timely filed.

Amendments to the Claims begin on page **2** of this paper.
A Summary of the February 5, 2020 Examiner Interview begins on page **8** of this paper.
A Summary of the April 3, 2020 Examiner Interview begins on page **10** of this paper.
Remarks begin on page **11** of this paper.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	:	Cold Spring Harbor Laboratory	
Inventors	:	James Hicks et al.	Examiner: Frank W. M. Lu
U.S. Serial No.	:	15/063,278	Art Unit: 1634
Filed	:	March 7, 2016	
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
New York, New York 10112
April 3, 2020

BY EFS
Commissioner for Patents
Alexandria, VA

FIFTH INFORMATION DISCLOSURE STATEMENT

In accordance with their duty of disclosure under 37 C.F.R. §1.56 and §1.97(a)-(d), applicants request that the documents listed hereunder and on the attached IDS form, hereto as **Exhibit A**, be considered and made of record in the above-identified application.

This Fifth Information Disclosure Statement lists documents filed during the continuing prosecution of a Notice of opposition to a European Patent filed on January 25, 2019 against counterpart foreign patent, European Patent No. EP2630263.

Reference item **1** is the Patentee's response to the notice of opposition filed August 8, 2019 in connection with EP2630263. Reference item **2** is a Summons to attend opposition oral proceedings issued by the European Patent Office on September 26, 2019 in connection with EP2630263.

Reference item **3** is the Provisional and non-binding opinion of the Opposition Division issued by the European Patent Office on September 26, 2019 concerning EP2630263. Reference item **4** is the Patentee's reply to the provisional and non-binding opinion of the Opposition

Division cited in reference item **3** filed November 7, 2019 concerning EP2630263.

Reference item **5** is a Submission in opposition proceedings filed by the Opponent on March 20, 2020 concerning EP2630263. Reference item **6** is cited in reference item **5.**

Copies of reference items **1-6** are attached hereto as **Exhibits 1-6**, respectively.

1. Patentee's response to the notice of opposition, filed August 8, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263 (**Exhibit 1**);

2. Summons to attend opposition oral proceedings, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263 (**Exhibit 2**);

3. Provisional and non-binding opinion of the Opposition Division, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263 (**Exhibit 3**);

4. Patentee's reply to the provisional and non-binding opinion of the Opposition Division, filed November 7, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263 (**Exhibit 4**);

5. Submission in opposition proceedings, filed March 20, 2020, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263 (**Exhibit 5**); and

6. Wang et al. (2002) "Digital Karyotyping", PNAS 99(25): 16156-16161 (**Exhibit 6**).

This Information Disclosure Statement is being submitted under 37 C.F.R. §1.97(c), *viz.* after the period specified in 37 C.F.R. §1.97(b) provided that the information disclosure statement is filed before the mailing date of any of a final action under 37 C.F.R. §1.113, a notice of allowance under 37 C.F.R. §1.311, or an action that otherwise closes prosecution in the application.

Accordingly, per 37 C.F.R. §1.704(d), this paper contains only an information disclosure statement in compliance with 37 C.F.R. §§1.704(c)(6), (c)(8), (c)(9), or (c)(10), and will thus not result in a reduction of any potential adjustment of the term of any patent issuing from this application under 37 C.F.R. §1.703(a)-(e).

Per 37 C.F.R. §1.97(c)(2) and 37 C.F.R. §1.17(p), the fee for submitting this Fifth Supplemental Information Disclosure Statement is TWO HUNDRED FORTY DOLLARS ($240.00), and the undersigned hereby authorizes charging this amount to Deposit Account No. 03-3125.

The Examiner is respectfully requested to make the above-listed references of record in the subject application by initialing and dating the attached forms, and returning copies of the initialed and dated forms to applicant's undersigned attorney.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicant's undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the $240.00 fee, is deemed necessary in connection with the filing of this Fifth Information Disclosure Statement. However, if any fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP
30 Rockefeller Plaza
New York, New York 10112
(212) 278-0400

Certificate of Transmittal

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

/Darren Haber/ April 3, 2020

Darren Haber Date

PTO/SB/08a (01-10)
Approved for use through 07/31/2012. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE

Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Not for submission under 37 CFR 1.99)

Application Number	15063278
Filing Date	2016-03-07
First Named Inventor	James Hicks
Art Unit	1634
Examiner Name	Frank W. M. Lu
Attorney Docket Number	81503-AA

U.S.PATENTS

Examiner Initial*	Cite No	Patent Number	Kind Code[1]	Issue Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
	1					

If you wish to add additional U.S. Patent citation information please click the Add button.

U.S.PATENT APPLICATION PUBLICATIONS

Examiner Initial*	Cite No	Publication Number	Kind Code[1]	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
	1					

If you wish to add additional U.S. Published Application citation information please click the Add button.

FOREIGN PATENT DOCUMENTS

Examiner Initial*	Cite No	Foreign Document Number[3]	Country Code[2] i	Kind Code[4]	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear	T[5]
	1							☐

If you wish to add additional Foreign Patent Document citation information please click the Add button

NON-PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc), date, pages(s), volume-issue number(s), publisher, city and/or country where published.	T[5]

INFORMATION DISCLOSURE STATEMENT BY APPLICANT (Not for submission under 37 CFR 1.99)		
	Application Number	15063278
	Filing Date	2016-03-07
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank W. M. Lu
	Attorney Docket Number	81503-AA

	1	Patentee's response to the notice of opposition, filed August 8, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263	☐
	2	Summons to attend opposition oral proceedings, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263	☐
	3	Provisional and non-binding opinion of the Opposition Division, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263	☐
	4	Patentee's reply to the provisional and non-binding opinion of the Opposition Division, filed November 7, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263	☐
	5	Submission in opposition proceedings, filed March 20, 2020, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263	☐
	6	Wang et al. (2002) "Digital Karyotyping", PNAS 99(25): 16156-16161	☐

If you wish to add additional non-patent literature document citation information please click the Add button

EXAMINER SIGNATURE

Examiner Signature		Date Considered	

*EXAMINER: Initial if reference considered, whether or not citation is in conformance with MPEP 609. Draw line through a citation if not in conformance and not considered. Include copy of this form with next communication to applicant.

[1] See Kind Codes of USPTO Patent Documents at www.USPTO.GOV or MPEP 901.04. [2] Enter office that issued the document, by the two-letter code (WIPO Standard ST.3). [3] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [4] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [5] Applicant is to place a check mark here if English language translation is attached.

GUARDANT HEALTH EX.1009.540

INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Not for submission under 37 CFR 1.99)

Application Number	15063278
Filing Date	2016-03-07
First Named Inventor	James Hicks
Art Unit	1634
Examiner Name	Frank W. M. Lu
Attorney Docket Number	81503-AA

CERTIFICATION STATEMENT

Please see 37 CFR 1.97 and 1.98 to make the appropriate selection(s):

☐ That each item of information contained in the information disclosure statement was first cited in any communication from a foreign patent office in a counterpart foreign application not more than three months prior to the filing of the information disclosure statement. See 37 CFR 1.97(e)(1).

OR

☐ That no item of information contained in the information disclosure statement was cited in a communication from a foreign patent office in a counterpart foreign application, and, to the knowledge of the person signing the certification after making reasonable inquiry, no item of information contained in the information disclosure statement was known to any individual designated in 37 CFR 1.56(c) more than three months prior to the filing of the information disclosure statement. See 37 CFR 1.97(e)(2).

☐ See attached certification statement.

☒ The fee set forth in 37 CFR 1.17 (p) has been submitted herewith.

☒ A certification statement is not submitted herewith.

SIGNATURE

A signature of the applicant or representative is required in accordance with CFR 1.33, 10.18. Please see CFR 1.4(d) for the form of the signature.

Signature	/Gary J. Gershik/	Date (YYYY-MM-DD)	2020-04-03
Name/Print	Gary J. Gershik	Registration Number	39,992

This collection of information is required by 37 CFR 1.97 and 1.98. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 1 hour to complete, including gathering, preparing and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

GUARDANT HEALTH EX.1009.541

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these record s.

2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.

3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.

4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).

5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.

6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).

7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.

8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.

9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

GUARDANT HEALTH EX.1009.542

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				
Extension-of-Time:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension - 3 months with $0 paid	2253	1	700	700
Miscellaneous:				
SUBMISSION- INFORMATION DISCLOSURE STMT	2806	1	120	120
		Total in USD ($)		**820**

Electronic Acknowledgement Receipt

EFS ID:	39063475
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	03-APR-2020
Filing Date:	07-MAR-2016
Time Stamp:	19:53:27
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$820
RAM confirmation Number	E202043J53440575
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		200403_81503_AA_Response_to_Non-Final_Office_Action_JTS.pdf	102317 f23f74fbacff20c3c7a2f95977cde64dc1be8bc6	yes	16

Multipart Description/PDF files in .zip description

Document Description	Start	End
Applicant Arguments/Remarks Made in an Amendment	11	16
Applicant summary of interview with examiner	10	10
Applicant summary of interview with examiner	8	9
Claims	2	7
Amendment/Req. Reconsideration-After Non-Final Reject	1	1

Warnings:

Information:

2	Transmittal Letter	200403_81503_AA_Transmittal_Letter_JTS.pdf	40740 61652e2373a5b392068b1e48ffae61a0d98b9a8a	no	4

Warnings:

Information:

3	Information Disclosure Statement (IDS) Form (SB08)	200403_81503_AA_Information_Disclosure_Statement_Form_Exhibit_A_JTS.pdf	69017 1c5af4dfd79eae7752e5ee01019356624595 10f1	no	4

Warnings:

Information:

This is not an USPTO supplied IDS fillable form

4	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_1_JTS.pdf	832605 ac4448c1472c3d3827d60f4c731aee61402ff2d1	no	18

Warnings:

GUARDANT HEALTH EX.1009.546

Information:					
5	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_2_JTS.pdf	40366 3761ec8f00b3991f0bc762c20c1621691ab720b6	no	1
Warnings:					
Information:					
6	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_3_JTS.pdf	200515 5d6ecf997a4480fb0286ba3bff53c21c30d517a2	no	4
Warnings:					
Information:					
7	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_4_JTS.pdf	183628 6b82dd84a69fb45dd00528e99add3ebeeabb3ae1	no	4
Warnings:					
Information:					
8	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_5_JTS.pdf	1921850 dd75806825f5c1b79ba360f71ea0e243475da555	no	14
Warnings:					
Information:					
9	Non Patent Literature	200403_81503_AA_NPL_Documents_Exhibit_6_JTS.pdf	1442142 9b5213b2740b53bb9c97c95a11db7113565b3ad5	no	6
Warnings:					
Information:					
10	Fee Worksheet (SB06)	fee-info.pdf	32649 e5dd7b396c7289b939e4f75009410998fc432487	no	2
Warnings:					
Information:					
			Total Files Size (in bytes):	4865829	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

REMARKS

Amendment to the Claims

By this Amendment, applicants have hereinabove canceled claims 1-87 and added claims 88-107. Support for the added claims may be found, *inter alia*, in the specification as filed as shown in the table below:

Claim	Specification
88	Page 15, lines 2-26; page 39, lines 12-13, page 43, lines 19-23, page 44, lines 4-7, page 44, lines 15-21; page 44, lines 27-29, page 51, lines 9-11; page 45, lines 11-14 and 28-33, page 61, lines 19-22 and lines 29-33
89	Page 15, line 27 to page 16, line 2, Page 45, lines 4-7
90	Page 16, lines 3-7
91	Page 16, lines 8-23, page 45, lines 28-33
92	Page 16, line 24 to page 17, line 8
93	Page 17, lines 11-16
94	Page 18, line 18 to page 19, line 29, page 44, lines 15-21; page 51, lines 9-11; page 45, lines 11-14 and 28-33
95	Page 24, lines 4-6
96	Page 24, lines 7-8 and page 51, lines 17-21
97	Page 23, lines 1-4
98	Page 23, lines 12-13
99	Page 24, lines 9-10
100	Page 39, lines 13-17
101	Page 24, lines 11-14
102	Page 24, lines 15-17
103	Page 24, lines 4-6, Page 29, lines 4-6, page 60, lines 21-23
104	Page 48, lines 17-18
105	Page 47, lines 20-23

106	Page 45, lines 9-10
107	Page 42, lines 15-17 and page 43, lines 12-15

Accordingly, upon entry of this Amendment, claims 88-107 are pending and under consideration in the subject application.

Rejection Under 35 U.S.C. § 101

In the October 3, 2019 Office Action, the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 101 as allegedly directed to a judicial exception (*i.e.*, a law of nature, a natural phenomenon, or an abstract idea) without significantly more.

The Examiner's specific rationale is set forth on pages 4-6 of the Office Action.

Applicant's response

In reply, applicant incorporates the Remarks of its April 17, 2019 Amendment, and maintains that the claimed invention amounts to significantly more than the alleged judicial exception at least because it a) provides an improvement over existing methods for determining copy number from genomic material, and b) requires physical processes.

The claims are patent-eligible under 35 U.S.C. § 101 in view of the January 2019 Revised Patent Subject Matter Eligibility Guidance and the October 2019 Update.

The U.S. Patent and Trademark Office has promulgated an October 2019 Patent Eligibility Guidance Update effective October 17, 2019 (hereinafter "October 2019 Update"). Federal Register, Vol. 84, No. 202, October 18, 2019, 55942-43. The January 2019 Guidance and the October 2019 Update make it clear that "Step 2A" of the *Alice/Mayo* test has two separate prongs. Prong One evaluates whether a claim recites a judicial exception by determining whether specific

limitations in the claim fall within the subject matter groupings of abstract ideas provided in the January 2019 Guidance. If the claim is deemed to recite a judicial exception, then Prong Two evaluates whether the judicial exception is integrated into a practical application. January 2019 Guidance at 54-55; *see* October 2019 Update at 10.

The identified additional elements need not individually be non-routine or non-conventional. *See* January 2019 Guidance at 55; *see also* October 2019 Update at 12-13. The new guidance explains that an invention that provides an improvement to a technology or technical field is indicative that the invention integrates the judicial exception into a practical application. *Id.* Further, the October 2019 Update makes clear that "[c]onsideration of improvements is relevant to the integration analysis regardless of the technology of the claimed invention." October 2019 Update at 13.

Finally, "Step 2B" evaluates whether the claim provides an inventive concept. January 2019 Guidance at 56; *see* October 2019 Update at 16. This is accomplished by evaluating whether the additional elements amount to significantly more than the judicial exception itself, i.e., the inventive concept. *Id.* An improvement to a technology or technical field indicates that an inventive concept is present. *Id.*

Applicant submits that the claims are patent-eligible under the January 2019 Guidance because, as stated above and explained below, the claimed invention provides an improvement which amounts to significantly more than the alleged judicial exception.

The claimed method to determine copy number from genomic material produces an improvement in the accuracy of calculating copy number information by eliminating unequal amplification of all molecules and thus provides a benefit over existing technology in the field by preventing amplification distortion. Consequently, the claimed method amounts to significantly more than the alleged judicial exception.

The January 2019 Guidance makes clear that an additional element or combination of elements that reflects an improvement to a technology or technical field is an "exemplary consideration[]" indicative that the additional element or combination of elements integrated the judicial exception into a practical application. *Id.* at 55; *see* October 2019 Update at 11. Further, an improvement to a technology or technical field indicates that an inventive concept is present.

The pending claims are directed to an improved method for assessing copy number of genomic material. By subjecting unique tagged nucleic acid molecules to amplification, the claimed method provides copy number information that is unaffected by amplification distortion. As discussed in the Specification, the claimed method is superior to other methods for ascertaining copy number by eliminating the influence of amplification distortion.

Applicant respectfully disagrees with the Examiner's assertion that independent claims 68 and 75, and dependent claims 69-72 and 82, fall within the mental process grouping of abstract ideas. Independent claim 68 recites a series of steps including (a)obtaining segments of genomic nucleic acids from a sample, and (b)generating unique tagged nucleic acid molecules. These are clearly not mental steps. Further, the polymerase chain reaction (PCR) in step (c) is not a mental step. The sequencing in step (d) is a physical step as well. Steps (e) and (f) are not divorced from these physical steps, but rather require that the physical steps be performed. Steps (e) and (f) can only be performed if the physical steps are performed, e.g. step (f) requires counting the number of "unique tagged nucleic acid molecules" which are physical molecules from steps (b) and (c) of claim 1. It is therefore improper to analyze steps (e) and (f) as purely mental steps.

As noted above, "Step 2B" of the *Alice/Mayo* test declares that the evaluation of additional elements of the claim must be considered when determining if said claim amounts to significantly more than the

judicial exception itself, i.e. whether the claim as a whole provides an inventive concept. January 2019 Guidance at 56; *see* October 2019 Update at 16.

Accordingly, the claims do not simply recite the alleged judicial exception without anything significantly more. As discussed above, the claims are directed to an improved method. Thus, the claims are eligible under the January 2019 Guidance and the October 2019 Update, and subsequently under 35 U.S.C. § 101.

In view of the foregoing remarks, applicant respectfully requests that the Examiner reconsider and withdraw the rejections under 35 U.S.C. § 101.

Rejection under 35 U.S.C. § 112(b) or § 112(pre-AIA), Second Paragraph

In the October 3, 2019 Office Action, the Examiner rejected claims 68-75 and 77-87 under 35 U.S.C. § 112(b) or 35 U.S.C. § 112, second paragraph, as allegedly indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The Examiner's specific rationale is set forth on pages 2-3 of the October 3, 2019 Office Action.

Applicant's Response

In response, applicant has proposed new claim language. Applicant submits that the amended claims are clear and respectfully requests that the Examiner reconsider and withdraw the rejections under 35 U.S.C. § 112(b) or 35 U.S.C. § 112, second paragraph.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee, other than the $700.00 fee for a Petition for a Three-month Extension of Time for a small entity, is deemed necessary in connection with the filing of this Amendment. However, if any fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant(s)
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

/Darren Haber/ April 3, 2020

Darren Haber Date

AMENDMENTS TO THE CLAIMS

Pursuant to 37 C.F.R. § 1.121(c), this listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

1-87. (Canceled)

88. (New) A method for obtaining copy number information from genomic material unaffected by amplification distortion, comprising:

a) obtaining segments of the genomic nucleic acids from a sample;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the unique tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each of the tag associated sequence reads of step (d); and

f) for a plurality of locations in step (e), counting the number of unique tagged nucleic acid molecules that have

been assigned to the same location on the genome to obtain a count for each location on the genome of the plurality of locations, thereby obtaining copy number information unaffected by amplification distortion.

89. (New) The method of claim 88, further comprising estimating a copy number of multiple locations within a region of the genome.

90. (New) The method of claim 88, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative copy number of the location.

91. (New) The method of claim 88, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative copy number of the first region of the genome to the copy number of the second region of the genome.

92. (New) The method of claim 91, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region

of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (j) for locations which comprise the third region.

93. (New) The method of claim 88, further comprising (l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative copy number of the region of the genome.

94. (New) A method for obtaining mRNA copy number information from mRNA transcripts, comprising:

a) generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase

reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) for a plurality of locations in step (e), counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, thereby obtaining a count for each location on the cDNA library of the plurality of locations on the cDNA library.

95. (New) The method of claim 88, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to PCR in

step (c) or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to sequencing in step (d).

96. (New) The method of claim 88, wherein each tagged nucleic acid molecule differs at more than one nucleotide.

97. (New) The method of claim 88, wherein tagging the segments to generate tagged nucleic acid molecules comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic material.

98. (New) The method of claim 88, wherein each tagged nucleic acid molecule comprises more than one tag.

99. (New) The method of claim 88, wherein the tags further comprise a sample tag.

100. (New) The method of claim 88, wherein a tagged nucleic acid molecule that differs by a single nucleotide is ignored when counting.

101. (New) The method of claim 88, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules of step (c).

102. (New) The method of claim 101, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

103. (New) The method of claim 95, wherein each of the unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array or on a microarray.

104. (New) The method of claim 88, for determining the proportion of

components in a mixture of genomic material.

105. (New) The method of claim 88, comprising comparing the copy number information obtained for different species to determine the relative count of those different species in a population.

106. (New) The method of claim 88, wherein genomic material is obtained from blood or other bodily fluids.

107. (New) The method of claim 88, wherein genomic material is obtained from tumor cell DNA in bodily fluid.

Summary of the February 5, 2020 Examiner Interview

On February 5, 2020 the undersigned conducted a Telephonic Examiner Interview with Examiner Frank W. Lu and his Supervisor, Examiner Nguyen. Dr. Vladimir Drozdoff, Chief Patent Counsel of the assignee, was also present. Applicant acknowledges with appreciation the courtesy that Examiner Lu and Examiner Nguyen extended during the February 5, 2020 Telephonic Examiner Interview.

During the February 5, 2020 interview, the Examiners and the undersigned discussed the rejections under 35 U.S.C. §§ 101 and 112 set forth in the October 3, 2019 Office Action.

In discussing the rejection under 35 U.S.C. § 101, the undersigned emphasized that the claimed invention is directed to a novel method for determining copy number from genomic material unaffected by amplification distortion. The undersigned pointed out that, contrary to assertions in the October 3, 2019 Office Action, steps a-d of claim 68 and steps a-c of claim 75 are not possible to execute mentally, as they require obtaining segments of genomic material, generating molecules, and carrying out PCR, each of which is a physical process. Examiner Nguyen proposed including the language "from a sample" when referencing genomic material (e.g. obtaining genomic material from a sample) to clarify that the genomic material is physical. The undersigned respectfully considered the suggestion and agreed to incorporate the phrase into the claims where applicable to reinforce that the claimed invention requires manipulation of physical material.

The undersigned also pointed out that the January 2019 Revised Patent Subject Matter Eligibility Guidance requires considering a claim as a whole, even when the claim is inclusive of abstract and/or conventional steps. The undersigned emphasized that the January 2019 Revised Patent Subject Matter Eligibility Guidance requires consideration of all steps in the claim together, and considering

whether the claim as a whole provides a solution or improvement. Examiner Nguyen suggested that the claim be amended to reflect the improvement the invention provides. The undersigned respectfully considered the suggestion and agreed to incorporate the phrase "unaffected by amplification distortion" into the claims, as presented herein.

Regarding the rejection under 35 U.S.C. § 112(b) or § 112(pre-AIA), second paragraph, Examiner Lu alleged that the indefiniteness issue of claims 68 and 75 arises from the vagueness of whether counting is performed at only one location or at a plurality of locations. The undersigned proposed new language for the claims that would indicate that counting is performed at a plurality of locations. Examiner Lu agreed to consider such amended claims.

Accordingly, applicant is filing this Amendment consistent with the discussion during the February 5, 2020 interview. In the Remarks, applicant provides a full response to each of the rejections set forth in the October 3, 2019 Office Action.

Summary of the April 3, 2020 Examiner Interview

On April 3, 2020 the undersigned conducted a Telephonic Examiner Interview with Examiner Frank W. Lu. Applicant acknowledges with appreciation the courtesy that Examiner Lu extended during the April 3, 2020 Telephonic Examiner Interview.

During the April 3, 2020 interview, the Examiner and the undersigned discussed similar issues as those discussed during the February 5, 2020 interview. The Examiner requested that the claims be clear that the claimed invention is not a series of mental steps, but rather requires manipulation of physical material.

Examiner Lu suggested that the language "on the genome" or "on the cDNA library" be added to claims 88(f) and 94(e), respectively. Examiner Lu explained that doing so would help advance the application toward allowance. The undersigned respectfully considered the suggestion and agreed to incorporate the above-mentioned phrases into the claims, as presented herein.

Accordingly, applicant is filing this Amendment consistent with the discussion during the April 3, 2020 interview. In the Remarks below, applicant provides a full response to each of the rejections set forth in the October 3, 2019 Office Action.

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☑ SMALL ☐ MICRO

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A		N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A		N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*		x $40 =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*		x $210 =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).				
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))					
* If the difference in column 1 is less than zero, enter "0" in column 2.				TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT	06/04/2020	CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0		x $50 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	*** 3	= 0		x $230 =	0
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	0

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT		CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=		x $0 =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=		x $0 =	
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.	SLIE
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".	/ANISSA D DEVLIN/
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".	
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.	

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
June 4, 2020

BY EFS
Commissioner for Patents
Alexandria, VA

SUPPLEMENTAL AMENDMENT

This Amendment supplements the Amendment filed by applicants on April 3, 2020 in response to the October 3, 2019 Non-Final Office Action issued by the United States Patent and Trademark Office in connection in connection with the above-identified application.

Please amend the subject application as follows:

Amendments to the Claims:

Pursuant to 37 C.F.R. §1.121(c), this listing of claims will replace all prior versions, and listings, of claims in the subject application:

Listing of Claims

1-87. (Canceled)

88. (Currently Amended) A method for obtaining copy number information from genomic nucleic acids in a sample ~~material unaffected by amplification distortion~~, comprising:

a) obtaining segments of the genomic nucleic acids from the [[a]] sample;

b) tagging the segments with nucleic acid tags, thereby generating unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the unique tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence associated with the genomic nucleic acids of each of the tag associated sequence reads of step (d); and

f) for a plurality of locations in step (e), counting the number of unique tagged nucleic acid molecules that have been assigned to the same location on the genome to obtain a count for each location on the genome of the plurality

of locations, thereby obtaining copy number information unaffected by amplification distortion.

89. (Previously Presented) The method of claim 88, further comprising estimating a copy number of multiple locations within a region of the genome.

90. (Previously Presented) The method of claim 88, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative copy number of the location.

91. (Previously Presented) The method of claim 88, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative copy number of the first region of the genome to the copy number of the second region of the genome.

92. (Previously Presented) The method of claim 91, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number

of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (j) for locations which comprise the third region.

93. (Previously Presented) The method of claim 88, further comprising (l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative copy number of the region of the genome.

94. (Currently Amended) A method for obtaining mRNA copy number information from mRNA transcripts in a sample, comprising:

a) generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts in the sample to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step

(ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) for a plurality of locations in step (e), counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, thereby obtaining a count for each location on the cDNA library of the plurality of locations on the cDNA library.

95. (Previously Presented) The method of claim 88, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to PCR in step (c) or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to sequencing in step (d).

96. (Previously Presented) The method of claim 88, wherein each tagged nucleic acid molecule differs at more than one nucleotide.

97. (Previously Presented) The method of claim 88, wherein tagging the segments to generate tagged nucleic acid molecules comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic material.

98. (Previously Presented) The method of claim 88, wherein each tagged nucleic acid molecule comprises more than one tag.

99. (Previously Presented) The method of claim 88, wherein the tags further comprise a sample tag.

100. (Previously Presented) The method of claim 88, wherein a tagged nucleic acid molecule that differs by a single nucleotide is ignored when counting.

101. (Previously Presented) The method of claim 88, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules of step (c).

102. (Previously Presented) The method of claim 101, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

103. (Previously Presented) The method of claim 95, wherein each of the unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array or on a microarray.

104. (Previously Presented) The method of claim 88, for determining the proportion of components in a mixture of genomic material.

105. (Previously Presented) The method of claim 88, comprising comparing the copy number information obtained for different species to determine the relative count of those different species in a population.

106. (Previously Presented) The method of claim 88, wherein genomic material is obtained from blood or other bodily fluids.

107. (Previously Presented) The method of claim 88, wherein genomic material is obtained from tumor cell DNA in bodily fluid.

REMARKS

Claims 88-107 are pending in the subject application. Pending claims 88 and 94 have been amended. Accordingly, claims 88-107 are still pending.

Summary of June 4, 2020 Telephonic Examiner Interview

On June 4, 2020 Examiner Frank W.M. Lu and the undersigned discussed claim amendments which may place the application in condition for allowance. No agreement was reached on June 4, 2020 other than the claim amendments presented in this Supplemental Amendment. Examiner Lu agreed to schedule a further Examiner interview to continue discussing potential additional claim amendments to place this application in condition for allowance. Applicants look forward to the further discussion with Examiner Lu.

If a telephone conference would be of assistance in advancing the prosecution of the subject application, applicant's undersigned attorneys invite the Examiner to telephone them at the number provided below.

No fee is deemed necessary in connection with the filing of this Supplemental Amendment. However, if any fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorney for Patentees
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

/Linsheng Li/ June 4, 2020

Linsheng Li Date

Electronic Acknowledgement Receipt

EFS ID:	39633367
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Linsheng Li
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	04-JUN-2020
Filing Date:	07-MAR-2016
Time Stamp:	21:19:29
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		200604_81503_AA_Supplemental_Amendment_JTS.pdf	109951 ab9e6c93e51e1da740f2ceb6dc451b48fa1b9a5f	yes	9

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Supplemental Response or Supplemental Amendment	1	1
	Claims	2	7
	Applicant Arguments/Remarks Made in an Amendment	8	9
Warnings:			
Information:			
	Total Files Size (in bytes):	109951	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 06/12/2020

COOPER & DUNHAM, LLP
30 Rockefeller Plaza
20th Floor
NEW YORK, NY 10112

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

NOTIFICATION DATE	DELIVERY MODE
06/12/2020	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

DUSPTO@cooperdunham.com
patcomm-in-grp@cooperdunham.com
pdocketing@cooperdunham.com

GUARDANT HEALTH EX.1009.576

Examiner-Initiated Interview Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

All participants (applicant, applicant's representative, PTO personnel):

(1) FRANK LU. (3) _____.

(2) Mr. Gary Gershik (Reg. No. 39,992). (4) _____.

Date of Interview: 03 June 2020.

Type: ☑ Telephonic ☐ Video Conference
☐ Personal [copy given to: ☐ applicant ☐ applicant's representative]

Exhibit shown or demonstration conducted: ☐ Yes ☐ No.
If Yes, brief description: _____.

Issues Discussed ☐ 101 ☐ 112 ☐ 102 ☐ 103 ☑ Others
(For each of the checked box(es) above, please describe below the issue and detailed description of the discussion)

Claim(s) discussed: 88-107.

Identification of prior art discussed: _____.

Substance of Interview
(For each issue discussed, provide a detailed description and indicate if agreement was reached. Some topics may include: identification or clarification of a reference or a portion thereof, claim interpretation, proposed amendments, arguments of any applied references etc...)

There was no agreement to be reached in the examiner's amendment. Mr. Gershik will file a supplemental amendment and ask for another interview with SPE..

Applicant recordation instructions: It is not necessary for applicant to provide a separate record of the substance of interview.

Examiner recordation instructions: Examiners must summarize the substance of any interview of record. A complete and proper recordation of the substance of an interview should include the items listed in MPEP 713.04 for complete and proper recordation including the identification of the general thrust of each argument or issue discussed, a general indication of any other pertinent matters discussed regarding patentability and the general results or outcome of the interview, to include an indication as to whether or not agreement was reached on the issues raised.

☐ Attachment

/FRANK W LU/ Primary Examiner, Art Unit 1634	June 3, 2020

GUARDANT HEALTH EX.1009.577

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☐ LARGE ☑ SMALL ☐ MICRO

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A		N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A		N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*		x $40 =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*		x $210 =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).				
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))					
* If the difference in column 1 is less than zero, enter "0" in column 2.				TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT	08/11/2020	CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 20	Minus	** 20	= 0		x $50 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	*** 3	= 0		x $230 =	0
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	0

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT		CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=		x $0 =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=		x $0 =	
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.	SLIE
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".	/SHARONE R MOORE/
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".	
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.	

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	James Hicks et al.	
U.S. Serial No.	:	15/063,278	Examiner: Frank W. M. Lu
Filing Date	:	March 7, 2016	Art Unit: 1634
Confirmation No.	:	4038	
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

30 Rockefeller Plaza
20th Floor
New York, New York 10112
August 11, 2020

BY EFS
Commissioner for Patents
Alexandria, VA

SECOND SUPPLEMENTAL AMENDMENT

This Amendment supplements the Supplemental Amendment filed by applicants on June 4, 2020 in response to the October 3, 2019 Non-Final Office Action issued by the United States Patent and Trademark Office in connection in connection with the above-identified application.

Please amend the subject application as follows:

Amendments to the Claims:

Pursuant to 37 C.F.R. §1.121(c), this listing of claims will replace all prior versions, and listings, of claims in the subject application:

Listing of Claims

1-87. (Canceled)

88. (Currently Amended) A method for obtaining copy number information from genomic nucleic acids in a sample, comprising:

a) obtaining segments of the genomic nucleic acids from the sample;

b) tagging the segments with nucleic acid tags, thereby generating a plurality of unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags;

c) subjecting the plurality of the unique tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a genome by mapping the subsequence ~~associated with the genomic nucleic acids~~ of each of the tag associated sequence reads of step (d) corresponding to a segment of the unique tagged nucleic acid molecule to a location of the genome; and

f) ~~for~~ at each of a plurality of locations corresponding to

a segment of a unique tagged nucleic acid molecule in step (e), counting the number of unique tagged nucleic acid molecules that have been assigned to the same location on the genome to obtain a count for each location on the genome of the plurality of locations, thereby obtaining copy number information from genomic nucleic acids in the sample ~~unaffected by amplification distortion~~.

89. (Previously Presented) The method of claim 88, further comprising estimating a copy number of multiple locations within a region of the genome.

90. (Previously Presented) The method of claim 88, further comprising comparing a count from the counts obtained in step (f) for a location on the genome to a count for the same location obtained from a reference sample, thereby estimating a relative copy number of the location.

91. (Previously Presented) The method of claim 88, further comprising

g) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a first region of the genome, wherein the first region comprises more than one location;

h) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a second region of the genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative copy number of the first region of the genome to the copy number of the second region of the genome.

92. (Previously Presented) The method of claim 91, wherein step (h) further comprises

j) summing the counts from the counts obtained in step (f) for locations on the genome which comprise a third region of the genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (j) for locations which comprise the third region.

93. (Previously Presented) The method of claim 88, further comprising (l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the genome; m) summing the counts obtained from a reference sample for the same region of the genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative copy number of the region of the genome.

94. (Currently Amended) A method for obtaining mRNA copy number information from mRNA transcripts in a sample, comprising:

a) generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts in the sample to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first

order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of at least one of steps (i) and (iii) comprise nucleic acid tags, such that each tagged nucleic acid molecule is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) ~~for~~ at each of a plurality of locations in step (e), counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned to the same location on the cDNA library, thereby obtaining a count for each location on the cDNA library of the plurality of locations on the cDNA library.

95. (Previously Presented) The method of claim 88, wherein the unique tagged nucleic acid molecules are subject to hybrid capture prior to PCR in step (c) or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to sequencing in step (d).

96. (Previously Presented) The method of claim 88, wherein each tagged nucleic acid molecule differs at more than one nucleotide.

97. (Previously Presented) The method of claim 88, wherein tagging the segments to generate tagged nucleic acid molecules comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic material.

98. (Previously Presented) The method of claim 88, wherein each tagged nucleic acid molecule comprises more than one tag.

99. (Previously Presented) The method of claim 88, wherein the tags further comprise a sample tag.

100. (Previously Presented) The method of claim 88, wherein a tagged nucleic acid molecule that differs by a single nucleotide is ignored when counting.

101. (Previously Presented) The method of claim 88, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules of step (c).

102. (Previously Presented) The method of claim 101, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

103. (Previously Presented) The method of claim 95, wherein each of the unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array or on a microarray.

104. (Previously Presented) The method of claim 88, for determining the proportion of components in a mixture of genomic material.

105. (Previously Presented) The method of claim 88, comprising comparing the copy number information obtained for different species to determine the relative count of those different species in a population.

106. (Previously Presented) The method of claim 88, wherein genomic material is obtained from blood or other bodily fluids.

107. (Previously Presented) The method of claim 88, wherein genomic material is obtained from tumor cell DNA in bodily fluid.

REMARKS

Claims 88-107 are pending in the subject application. Pending claims 88 and 94 have been amended. Accordingly, claims 88-107 are still pending.

Summary of August 5, 2020 Telephonic Examiner Interview

During the August 5, 2020 Interview, Examiner Frank W.M. Lu and the undersigned discussed claim amendments which may place the application in condition for allowance. Examiner Lu requested that the aforementioned claim amendments be formally filed for consideration.

If a telephone conference would be of assistance in advancing the prosecution of the subject application, applicant's undersigned attorneys invite the Examiner to telephone them at the number provided below.

No fee is deemed necessary in connection with the filing of this Supplemental Amendment. However, if any fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorney for Patentees
Cooper & Dunham LLP
30 Rockefeller Plaza
20th Floor
New York, New York 10112
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on the date below.

/Darren Haber/ August 11, 2020

Darren Haber Date

Electronic Acknowledgement Receipt

EFS ID:	40252070
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	11-AUG-2020
Filing Date:	07-MAR-2016
Time Stamp:	14:56:17
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		200811_81503_AA_Second_Supplemental_Amendment_JTS.pdf	104181 a09a7899d412011ab675889d92f4075851cb5cfc	yes	9

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Supplemental Response or Supplemental Amendment	1	1
	Claims	2	7
	Applicant Arguments/Remarks Made in an Amendment	8	9
Warnings:			
Information:			
	Total Files Size (in bytes):	104181	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 10/16/2020

COOPER & DUNHAM LLP
845 Third Avenue, 6th Floor
NEW YORK, NY 10022

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

NOTIFICATION DATE	DELIVERY MODE
10/16/2020	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

DUSPTO@cooperdunham.com
patcomm-in-grp@cooperdunham.com
pdocketing@cooperdunham.com

GUARDANT HEALTH EX.1009.590

Office Action Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☑ Responsive to communication(s) filed on 4/3/2020, 6/4/2020, and 8/11/2020.

☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2a)☑ This action is **FINAL.** 2b) ☐ This action is non-final.

3)☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

4)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

5) ☑ Claim(s) 88-107 is/are pending in the application.

5a) Of the above claim(s) _____ is/are withdrawn from consideration.

6) ☐ Claim(s) _____ is/are allowed.

7) ☑ Claim(s) 88-107 is/are rejected.

8) ☐ Claim(s) _____ is/are objected to.

9) ☐ Claim(s) _____ are subject to restriction and/or election requirement

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to **PPHfeedback@uspto.gov.**

Application Papers

10)☐ The specification is objected to by the Examiner.

11)☑ The drawing(s) filed on 4/17/2019 is/are: a)☑ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

a)☐ All b)☐ Some** c)☐ None of the:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)

2) ☑ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b) Paper No(s)/Mail Date 4/3/2020.

3) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date _____.

4) ☐ Other: _____.

DETAILED ACTION

Response to Amendment

1. Applicant's responses to the office action filed on April 3, 2020, June 4, 2020, and August 11, 2020 have been entered. The claims pending in this application are claims 88-107. Rejection and/or objection not reiterated from the previous office action are hereby withdrawn in view of applicant's amendment filed on April 3, 2020, June 4, 2020, and August 11, 2020. Claims 88-107 will be examined.

Claim Objections

2. Claim 88 is objected to because of the following informality: "each location on the genome of the plurality of locations" in step f) should be "each location of the plurality of locations on the genome".

3. Claim 94 is objected to because of the following informalities: (1) "each tagged nucleic acid molecule" in step a) should be "each of tagged nucleic acid molecules"; and (2) "each location on the cDNA library" in step e) should be "each location".

4. Claim 96 or 98 is objected to because of the following informality: "each tagged nucleic acid molecule" in step a) should be "each of the tagged nucleic acid molecules".

5. Claim 97 is objected to because of the following informality: "tagging the segments to generate tagged nucleic acid molecules" should be "said tagging the segments with nucleic acid tags".

6. Claim 100 is objected to because of the following informality: "when counting" should be "during the process of said counting the number of unique tagged nucleic acid molecules that have been assigned to the same location on the genome".

7. Claim 105 is objected to because of the following informalities: (1) "obtained for different species" should be "obtained from different species"; and (2) "those different species" should be "the different species".

Appropriate correction is required.

Claim Rejections - 35 USC § 112

8. The following is a quotation of 35 U.S.C. 112(b):

> (b) CONCLUSION.—The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the inventor or a joint inventor regards as the invention.

> The following is a quotation of 35 U.S.C. 112 (pre-AIA), second paragraph:
> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

9. Claims 88-107 are rejected under 35 U.S.C. 112(b) or 35 U.S.C. 112 (pre-AIA), second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which the inventor or a joint inventor (or for applications subject to pre-AIA 35 U.S.C. 112, the applicant), regards as the invention.

10. Claim 88 is rejected as vague and indefinite in view of the preamble and step f) because it is unclear that copy number information of which specific nucleic acid is obtained from genomic nucleic acids in a sample. Please clarify.

11. Claim 88 is rejected as vague and indefinite in view of step b) because it is unclear what means "corresponding to". Does the phrase "a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags" mean that "a segment of the segments of the genomic nucleic acids from step (a) and a nucleic acid tag of the nucleic acid tags"? Please clarify.

12. Claim 88 is rejected as vague and indefinite in view of step e) because it is unclear how the subsequence of each of the tag associated sequence reads of step (d) corresponds to a segment of the unique tagged nucleic acid molecule. Please clarify.

13. Claim 88 recites the limitation "the unique tagged nucleic acid molecule" in step e) of the claim. There is insufficient antecedent basis for this limitation in the claim because step c) only has a phrase "unique tagged nucleic acid molecules". Please clarify.

14. Claim 88 is rejected as vague and indefinite in view of step f) because step f) does not have each of a plurality of locations corresponding to a segment of a unique tagged nucleic acid molecule in step (e). Furthermore, since one copy of genomic nucleic acids can be cleaved to multiple copies of segments, although step b) requires that each of the unique tagged nucleic acid molecules comprises a portion corresponding to a segment of the segments of the genomic nucleic acids from step (a) and a portion corresponding to a nucleic acid tag of the nucleic acid tags, without knowing that one copy of genomic nucleic acids is cleaved to how many copies of segments, it is unclear why copy number information from genomic nucleic acids in the sample can be obtained by only counting the number of unique tagged nucleic acid molecules that have been assigned to the same location on the genome and obtaining a count for each location of the plurality of locations on the genome. Please clarify.

15. Claim 89 is rejected as vague and indefinite because it is unclear that a copy number of what of multiple locations within a region of the genome is estimated. Please clarify.

16. Claim 90 or 91 recites the limitation "the counts obtained in step (f) for a location on the genome" in the claim. There is insufficient antecedent basis for this limitation in the claim because there is no word "counts" in step (f) of claim 88. Please clarify.

17. Claim 91 is rejected as vague and indefinite. Since the genome in the claim includes a first region of the genome and a second region of the genome, each of step g) and step h) can be reasonably read as summing the counts from the counts obtained in step (f) for locations on the genome and step g) and step h) can be considered as an identical step such that the value obtained in step (g) is identical to the value obtained in step (h). Since the counts in step g) is not related to the first region of the genome and the counts in step h) is not related to the second region of the genome, the relative copy number of the first region of the genome to the copy number of the second region of the genome cannot be estimated by comparing the value obtained in step (g) to the value obtained in step (h). Please clarify.

18. Claim 94 is rejected as vague and indefinite in view of the preamble because it is unclear that mRNA copy number information of what specific mRNA is obtained from mRNA transcripts in a sample. Furthermore, although the claim is directed to a method for obtaining mRNA copy number information from mRNA transcripts in a sample while step e) of the claim is required for obtaining a count for each location on the cDNA library of the plurality of locations on the cDNA library, the preamble and the last method step do not correspond each other. Please clarify.

19. Claim 94 is rejected as vague and indefinite in view of step a). Since step a) does not require that nucleic acid tags of the primers of at least one of steps (i) and (iii) are different, it is unclear why each of tagged nucleic acid molecules must be unique. Furthermore, if the phrase "the primers of at least one of steps (i) and (iii) comprise nucleic acid tags" is read as "the primers of one of steps (i) and (iii) comprise nucleic acid tags", only one of the first order derivative strands and the second order derivative strands has nucleic acid tags and it is unclear

why both the first order derivative strands and the second order derivative strands can be the tagged nucleic acid molecules. Please clarify.

20. Claim 94 is rejected as vague and indefinite in view of step e) because the phrase "at each of a plurality of locations in step (e)" does not make sense since step e) does not mention a plurality of locations before this phrase. Furthermore, it is unclear what is the relationship between nucleic acid tags in step a) and a different tag that have been assigned to the same location on the cDNA library in step e). Please clarify.

21. Claim 95 is rejected as vague and indefinite because it is unclear what means "are subject to hybrid capture". Please clarify.

22. Claim 97 recites the limitation "the genomic material" in the claim. There is insufficient antecedent basis for this limitation in the claim because there is no phrase "a genomic material" in claim 88. Please clarify.

23. Claim 100 is rejected as vague and indefinite because it is unclear what is the relationship between the tagged nucleic acid molecules in claim 88 and a tagged nucleic acid molecule in claim 100. Please clarify.

24. Claim 104 is rejected as vague and indefinite because it is unclear that the proportion of what components is determined in a mixture of genomic material. Please clarify.

25. Claim 105 is rejected as vague and indefinite because it is unclear what is compared with the copy number information of what obtained from different species to determine the relative count of the different species in a population. Please clarify.

26. Claim 106 is rejected as vague and indefinite. Since "wherein" phrase is used to define a word or phrase which appears before and there is no phrase "genomic material" in claim 88, it is unclear why genomic material can be obtained from blood or other bodily fluids. Please clarify.

27. Claim 107 is rejected as vague and indefinite. Since "wherein" phrase is used to define a word or phrase which appears before and there is no phrase "genomic material" in claim 88, it is unclear why genomic material can be obtained from tumor cell DNA in bodily fluid. Please clarify.

Claim Rejections - 35 USC § 101

28. 35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

29. Claims 88-107 are rejected under 35 U.S.C. 101 because the claimed invention is directed to a judicial exception (i.e., a law of nature, a natural phenomenon, or an abstract idea) without significantly more.

The processes of claims 88-93 and 95-107 are directed to a process of obtaining copy number information from genomic nucleic acids in a sample wherein steps e) and f) in independent claim 88 and dependent claims 89-93, 102, and 105 fall within the mental process grouping of abstract ideas. The process of claim 94 is directed to a process of obtaining mRNA copy number information from mRNA transcripts in a sample wherein steps d) and e) in the claim 94 fall within the mental process grouping of abstract ideas. **Note that steps in a claim that recite the manipulation of information through a series of mental steps would be considered a single abstract idea for purposes of analysis** (see MPEP 2106.04).

This judicial exception, the abstract idea, is not integrated into a practical application because the additional elements, steps e) and f) in independent claim 88, dependent claims 89-93, 102, and 105 and steps d) and e) in independent claim 94 are mental steps that do not integrate the recited judicial exception into a practical application. In a case that applicant

argues that mapping the subsequence of each of the tag associated sequence reads of step (d) corresponding to a segment of the unique tagged nucleic acid molecule to a location of the genome in step e) of claim 88 and mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library in step d) of claim 94 must be performed using a computer, the claimed subject matter still comprises an abstract idea that is computer mediated since the mapping steps in claims 88 and 94 can be performed using a generic computer. **Performing an abstract idea on a generic computer is not an additional element that is significantly more than the abstract idea itself**. *In Alice Corp. Pty. Ltd. v. CLS Bank Int'l* (573 U.S. 134 S. Ct. 2347, 110 USPQ2d 1976 (2014)) the Supreme Court stated regarding claims directed to an abstract idea:

> These cases demonstrate that the mere recitation of a generic computer cannot transform a patent-ineligible abstract idea into a patent-eligible invention. Stating an abstract idea "while adding the words 'apply it'" is not enough for patent eligibility. *Mayo*, *supra,* at ___ (slip op., at 3). Nor is limiting the use of an abstract idea "'to a particular technological environment.'" *Bilski*, *supra,* at 610–611. Stating an abstract idea while adding the words "apply it with a computer" simply combines those two steps, with the same deficient result. Thus, if a patent's recitation of a computer amounts to a mere instruction to "implemen[t]" an abstract idea "on . . . a computer," *Mayo*, *supra,* at ___ (slip op., at 16), that addition cannot impart patent eligibility. This conclusion accords with the pre-emption concern that undergirds our §101 jurisprudence. Given the ubiquity of computers, see 717 F. 3d, at 1286 (Lourie, J., concurring), wholly generic computer implementation is not generally the sort of "additional featur[e]" that provides any "practical assurance that the process is more than a drafting effort designed to monopolize the [abstract idea] itself." *Mayo*, 566 U. S., at ___ (slip op., at 8–9).

Regarding claims to computers and computer-readable media that execute an abstract idea, the court further stated:

> Petitioner's claims to a computer system and a computer-readable medium fail for substantially the same reasons. Petitioner conceded below that its media claims rise or fall with its method claims. En Banc Response Brief

for Defendant-Appellant in No. 11–1301 (CA Fed.) p. 50, n. 3. As to its system claims, petitioner emphasizes that those claims recite "specific hardware" configured to perform "specific computerized functions." Brief for Petitioner 53. But what petitioner characterizes as specific hardware-a "data processing system" with a "communications controller" and "data storage unit," for example, see App. 954,958, 1257-is purely functional and generic. Nearly every computer will include a "communications controller" and "data storage unit" capable of performing the basic calculation, storage, and transmission functions required by the method claims. See 717 F. 3d, at 1290 (Lourie, J., concurring). As a result, none of the hardware recited by the system claims "offers a meaningful limitation beyond generally linking 'the use of the [method] to a particular technological environment,' that is, implementation via computers." *Id.*, at 1291 (quoting *Bilski*, 561 U. S., at 610–611). Put another way, the system claims are no different from the method claims in substance. The method claims recite the abstract idea implemented on a generic computer; the system claims recite a handful of generic computer components configured to implement the same idea. This Court has long "warn[ed] . . . against" interpreting § 101 "in ways that make patent eligibility 'depend simply on the draftsman's art.'" *Mayo*, *supra*, at ___ (slip op., at 3) (quoting *Flook*, 437 U. S., at 593); see *id.*, at 590 ("The concept of patentable subject matter under § 101 is not 'like a nose of wax which may be turned and twisted in any direction . . . '"). Holding that the system claims are patent eligible would have exactly that result.

Because petitioner's system and media claims add nothing of substance to the underlying abstract idea, we hold that they too are patent ineligible under § 101.

The claims do not include additional elements that are sufficient to amount to significantly more than the judicial exception because independent claim 88, the steps a) to d) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see claims 1-21 of US Patent No. 9,670,536 B2 with a priority date of September 21, 2010, specially see step (a) and (b) of claim 17, and pages 119 and 120, online methods and Figure 1 from Hiatt *et al.,* Nature Methods, 7, 119-122, published online on January 17, 2010) while, in independent claim 94, the steps a) to c) are common, well-understood, routine and conventional methods used in the laboratory and are well known in the art (for example, see Figures 1a, 1c and 1d in page 529 and online methods from Plessy *et al.,*

Nature Methods, 7, 528-534, published online on June 13, 2010 wherein template-switching oligonucleotide in Figures 1a in page 529 is considered as a polynucleotide tail in ii) of step a) of claim 94 and the PCR primers of Figure 1d in page 529 are considered to comprise nucleic acid tags). The conventional steps do not comprise an inventive concept that transforms the claimed abstract ideas into a patent-eligible application of the abstract ideas. The claimed subject matters of claims 88-107 are processes directed to abstract ideas. When recited at this high level of generality, there is no meaningful limitation, such as a particular or unconventional machine or a transformation of a particular article, in this step that distinguishes it from well-understood, routine, and conventional data gathering activity engaged in by scientists prior to applicant's invention, and at the time the application was filed, *e.g.*, the routine and conventional techniques for performing steps a) to d) of claim 88 or steps a) to c) of claim 94. Further, it is well established that the mere physical or tangible nature of additional elements such as steps a) to d) of claim 88 or steps a) to c) of claim 94 do not automatically confer eligibility on a claim directed to an abstract idea (*see, e.g.*, *Alice Corp. v. CLS Bank Int'l*, 134 S. Ct. 2347, 2358-59 (2014)). Therefore, the claimed subject matters of claims 88-107 are without limitations that indicate that the claims are no different from the process claims directed to abstract ideas and are patent-ineligible.

Response to Arguments

30. Applicant's arguments with respect to claims 68-75 and 77-87 have been considered but are moot because the new ground of rejection does not rely on the prior rejection of record for any teaching or matter specifically challenged in the argument.

Conclusion

31. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

32. No claim is allowed.

33. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993)(See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/FRANK W LU/
Primary Examiner, Art Unit 1634
October 11, 2020

Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	Art Unit
	FRANK W LU	1634

CPC - Searched*		
Symbol	Date	Examiner
Update	10/11/2020	/FL/

CPC Combination Sets - Searched*		
Symbol	Date	Examiner
Update	10/11/2020	/FL/

US Classification - Searched*			
Class	Subclass	Date	Examiner
Update		10/11/2020	/FL/

* See search history printout included with this form or the SEARCH NOTES box below to determine the scope of the search.

Search Notes		
Search Notes	Date	Examiner
Update (see attached file)	10/11/2020	/FL/

Interference Search			
US Class/CPC Symbol	US Subclass/CPC Group	Date	Examiner

/FRANK LU/ Primary Examiner, Art Unit 1634	October 11, 2020

GUARDANT HEALTH EX.1009.603

Doc code: IDS
Doc description: Information Disclosure Statement (IDS) Filed

PTO/SB/08a (01-10)
Approved for use through 07/31/2012. OMB 0651-0031
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

INFORMATION DISCLOSURE STATEMENT BY APPLICANT

(Not for submission under 37 CFR 1.99)

Application Number	15063278
Filing Date	2016-03-07
First Named Inventor	James Hicks
Art Unit	1634
Examiner Name	Frank W. M. Lu
Attorney Docket Number	81503-AA

U.S.PATENTS

Examiner Initial*	Cite No	Patent Number	Kind Code[1]	Issue Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
	1					

If you wish to add additional U.S. Patent citation information please click the Add button.

U.S.PATENT APPLICATION PUBLICATIONS

Examiner Initial*	Cite No	Publication Number	Kind Code[1]	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
	1					

If you wish to add additional U.S. Published Application citation information please click the Add button.

FOREIGN PATENT DOCUMENTS

Examiner Initial*	Cite No	Foreign Document Number[3]	Country Code[2] i	Kind Code[4]	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear	T[5]
	1							☐

If you wish to add additional Foreign Patent Document citation information please click the Add button

NON-PATENT LITERATURE DOCUMENTS

Examiner Initials*	Cite No	Include name of the author (in CAPITAL LETTERS), title of the article (when appropriate), title of the item (book, magazine, journal, serial, symposium, catalog, etc), date, pages(s), volume-issue number(s), publisher, city and/or country where published.	T[5]

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

GUARDANT HEALTH EX.1009.604

INFORMATION DISCLOSURE STATEMENT BY APPLICANT (Not for submission under 37 CFR 1.99)		
	Application Number	15063278
	Filing Date	2016-03-07
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank W. M. Lu
	Attorney Docket Number	81503-AA

	1	Patentee's response to the notice of opposition, filed August 8, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263	☐
	2	Summons to attend opposition oral proceedings, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263	☐
	3	Provisional and non-binding opinion of the Opposition Division, issued by the European Patent Office on September 26, 2019, concerning European Patent No. EP2630263	☐
	4	Patentee's reply to the provisional and non-binding opinion of the Opposition Division, filed November 7, 2019, by Patentee, Cold Spring Harbor Laboratory, with the European Patent Office, concerning European Patent No. EP2630263	☐
	5	Submission in opposition proceedings, filed March 20, 2020, by Opponent, James Poole Limited, with the European Patent Office, concerning European Patent No. EP2630263	☐
	6	Wang et al. (2002) "Digital Karyotyping", PNAS 99(25): 16156-16161	☐

If you wish to add additional non-patent literature document citation information please click the Add button

EXAMINER SIGNATURE

Examiner Signature	/FRANK W LU/	Date Considered	10/11/2020

*EXAMINER: Initial if reference considered, whether or not citation is in conformance with MPEP 609. Draw line through a citation if not in conformance and not considered. Include copy of this form with next communication to applicant.

[1] See Kind Codes of USPTO Patent Documents at www.USPTO.GOV or MPEP 901.04. [2] Enter office that issued the document, by the two-letter code (WIPO Standard ST.3). [3] For Japanese patent documents, the indication of the year of the reign of the Emperor must precede the serial number of the patent document. [4] Kind of document by the appropriate symbols as indicated on the document under WIPO Standard ST.16 if possible. [5] Applicant is to place a check mark here if English language translation is attached.

ALL REFERENCES CONSIDERED EXCEPT WHERE LINED THROUGH. /F.W.L/

GUARDANT HEALTH EX.1009.605

Web Search History

date, time	web site	search string
10/12/2020 12:05:46 AM	Google	(tag or adaptor or adapter or linker) and ("nucleic acid" or polynucleotide or DNA) and (amplify or amplifying or amplification) and sequence read
10/12/2020 12:06:03 AM	Google Scholar	(tag or adaptor or adapter or linker) and ("nucleic acid" or polynucleotide or DNA) and (amplify or amplifying or amplification) and sequence read

EAST Search History

EAST Search History (Prior Art)

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L3	29	(tag or adaptor or adapter or linker) near10 ("nucleic acid" or polynucleotide or DNA) near20 (amplify or amplifying or amplification) near20 sequence near read and (PCR near (polymerase near chain near reaction)) same (amplify or amplifying or amplification)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	OFF	2020/10/12 00:02
L4	29	3 AND ((C12Q2535/122 OR C12Q1/683 OR C12Q2525/191 OR C12Q2537/16 OR C12Q2525/155 OR C12Q2537/159 OR C12Q1/6869 OR C12Q1/6874 OR C12Q1/6886 OR C12Q2600/156 OR C12Q1/6881 OR C12Q2537/165 OR C12Q1/6806 OR C12Q1/689 OR C12Q2563/179 OR C12Q2525/15 OR C12Q2525/197 OR C12Q2527/137 OR C12Q2545/113 OR C12Q2561/113 OR C12Q2563/131 OR C12Q2563/149 OR C12Q2565/50 OR C12Q2565/514 OR C12Q1/6809 OR C12Q1/6851 OR C12Q2600/158 OR C12Q2545/114 OR G16B25/00 OR G16B30/00 OR G16B30/20 OR G16B50/00 OR G16B35/10 OR C12N15/1065 OR C12N15/1093 OR C40B50/06).CPC.)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	OFF	2020/10/12 00:03

10/12/2020 12:04:14 AM
C:\Users\flu\Documents\EAST\Workspaces\~auto.02042018.005450.wsp

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 11/20/2020

COOPER & DUNHAM LLP
845 Third Avenue, 6th Floor
NEW YORK, NY 10022

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

NOTIFICATION DATE	DELIVERY MODE
11/20/2020	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

DUSPTO@cooperdunham.com
patcomm-in-grp@cooperdunham.com
pdocketing@cooperdunham.com

GUARDANT HEALTH EX.1009.608

Examiner-Initiated Interview Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

All participants (applicant, applicant's representative, PTO personnel):

(1) FRANK LU.

(2) Dave Nguyen (SPE Art Unit 1634).

(3) Gary Gershik (Reg. No. 39,992).

(4) Vladimir Drozdoff, Ph.D. and Darren Haber..

Date of Interview: 17 November 2020.

Type: ☑ Telephonic ☐ Video Conference
☐ Personal [copy given to: ☐ applicant ☐ applicant's representative]

Exhibit shown or demonstration conducted: ☐ Yes ☐ No.
If Yes, brief description: _____.

Issues Discussed ☑ 101 ☑ 112 ☐ 102 ☐ 103 ☐ Others
(For each of the checked box(es) above, please describe below the issue and detailed description of the discussion)

Claim(s) discussed: 88-107.

Identification of prior art discussed: _____.

Substance of Interview
(For each issue discussed, provide a detailed description and indicate if agreement was reached. Some topics may include: identification or clarification of a reference or a portion thereof, claim interpretation, proposed amendments, arguments of any applied references etc...)

Applicant's representatives and examiners discussed the rejections and possible amendments. The examiners agreed to withdraw 101 rejection on claims 88-107 in light of the teachings of the specification..

Applicant recordation instructions: It is not necessary for applicant to provide a separate record of the substance of interview.

Examiner recordation instructions: Examiners must summarize the substance of any interview of record. A complete and proper recordation of the substance of an interview should include the items listed in MPEP 713.04 for complete and proper recordation including the identification of the general thrust of each argument or issue discussed, a general indication of any other pertinent matters discussed regarding patentability and the general results or outcome of the interview, to include an indication as to whether or not agreement was reached on the issues raised.

☐ Attachment

/FRANK W LU/ Primary Examiner, Art Unit 1634	November 17, 2020

GUARDANT HEALTH EX.1009.609

PTO/SB/06 (09-11)
Approved for use through 1/31/2014. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

PATENT APPLICATION FEE DETERMINATION RECORD Substitute for Form PTO-875	Application or Docket Number 15/063,278	Filing Date 03/07/2016	☐ To be Mailed

ENTITY: ☑ LARGE ☐ SMALL ☐ MICRO

APPLICATION AS FILED - PART I

FOR	(Column 1) NUMBER FILED	(Column 2) NUMBER EXTRA		RATE ($)	FEE ($)
☐ BASIC FEE (37 CFR 1.16(a), (b), or (c))	N/A	N/A		N/A	
☐ SEARCH FEE (37 CFR 1.16(k), (i), or (m))	N/A	N/A		N/A	
☐ EXAMINATION FEE (37 CFR 1.16(o), (p), or (q))	N/A	N/A		N/A	
TOTAL CLAIMS (37 CFR 1.16(i))	minus 20 =	*		x $80 =	
INDEPENDENT CLAIMS (37 CFR 1.16(h))	minus 3 =	*		x $420 =	
☐ APPLICATION SIZE FEE (37 CFR 1.16(s))	If the specification and drawings exceed 100 sheets of paper, the application size fee due is $310 ($155 for small entity) for each additional 50 sheets or fraction thereof. See 35 U.S.C. 41(a)(1)(G) and 37 CFR 1.16(s).				
☐ MULTIPLE DEPENDENT CLAIM PRESENT (37 CFR 1.16(j))					
* If the difference in column 1 is less than zero, enter "0" in column 2.				TOTAL	

APPLICATION AS AMENDED - PART II

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT	11/25/2020	CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	* 19	Minus	** 20	= 0		x $100 =	0
	Independent (37 CFR 1.16(h))	* 2	Minus	*** 3	= 0		x $480 =	0
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	0

		(Column 1)		(Column 2)	(Column 3)			
AMENDMENT		CLAIMS REMAINING AFTER AMENDMENT		HIGHEST NUMBER PREVIOUSLY PAID FOR	PRESENT EXTRA		RATE ($)	ADDITIONAL FEE ($)
	Total (37 CFR 1.16(i))	*	Minus	**	=		x $0 =	
	Independent (37 CFR 1.16(h))	*	Minus	***	=		x $0 =	
	☐ Application Size Fee (37 CFR 1.16(s))							
	☐ FIRST PRESENTATION OF MULTIPLE DEPENDENT CLAIM (37 CFR 1.16(j))							
							TOTAL ADD'L FEE	

* If the entry in column 1 is less than the entry in column 2, write "0" in column 3.	LIE
** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 20, enter "20".	/WILLIAM N PHILLIPS/
*** If the "Highest Number Previously Paid For" IN THIS SPACE is less than 3, enter "3".	
The "Highest Number Previously Paid For" (Total or Independent) is the highest number found in the appropriate box in column 1.	

This collection of information is required by 37 CFR 1.16. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

If you need assistance in completing the form, call 1-800-PTO-9199 and select option 2.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s) : Cold Spring Harbor Laboratory

U.S. Serial No. : 15/063,278 Examiner : Frank W. M. Lu

Filing Date : March 7, 2016 Art Unit : 1634

Confirmation No. : 4038

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

845 Third Avenue
6th Floor
New York, NY 10022
November 25, 2020

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

AMENDMENT UNDER 37 C.F.R. §1.116 IN RESPONSE TO OCTOBER 16, 2020 FINAL OFFICE ACTION, REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0, AND SUMMARY OF NOVEMBER 17 AND 24, 2020 EXAMINER INTERVIEWS

This Amendment is a submission under 37 C.F.R. § 1.116 accompanying a Request for Consideration under the After Final Consideration Pilot Program 2.0 and responds to the issues raised in a October 24, 2020 Final Office Action issued in connection with the above-identified application. A response to the October 24, 2020 Final Office Action is due January 24, 2021. Accordingly, this Amendment is being timely filed.

Amendments to the Claims begin on page **2** of this paper.

A **Summary of November 17, 2020 Examiner Interview** begins on page **9** of this paper. A **Summary of November 24, 2020 Examiner Interview** begins on page **10** of this paper.

Remarks begin on page **11** of this paper.

Doc Code: A.NE.AFCP
Document Description: After Final Consideration Pilot Program Request

PTO/SB/434 (05-13)

CERTIFICATION AND REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0

Practitioner Docket No.:	Application No.:	Filing Date:
81503-AA/GJG/DH	15/063,278	March 7, 2016
First Named Inventor:	**Title:**	
James Hicks	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

APPLICANT HEREBY CERTIFIES THE FOLLOWING AND REQUESTS CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0 (AFCP 2.0) OF THE ACCOMPANYING RESPONSE UNDER 37 CFR 1.116.

1. The above-identified application is (i) an original utility, plant, or design nonprovisional application filed under 35 U.S.C. 111(a) [a continuing application (*e.g.*, a continuation or divisional application) is filed under 35 U.S.C. 111(a) and is eligible under (i)], or (ii) an international application that has entered the national stage in compliance with 35 U.S.C. 371(c).
2. The above-identified application contains an outstanding final rejection.
3. Submitted herewith is a response under 37 CFR 1.116 to the outstanding final rejection. The response includes an amendment to at least one independent claim, and the amendment does not broaden the scope of the independent claim in any aspect.
4. This certification and request for consideration under AFCP 2.0 is the only AFCP 2.0 certification and request filed in response to the outstanding final rejection.
5. Applicant is willing and available to participate in any interview requested by the examiner concerning the present response.
6. This certification and request is being filed electronically using the Office's electronic filing system (EFS-Web).
7. Any fees that would be necessary consistent with current practice concerning responses after final rejection under 37 CFR 1.116, *e.g.*, extension of time fees, are being concurrently filed herewith. [There is no additional fee required to request consideration under AFCP 2.0.]
8. By filing this certification and request, applicant acknowledges the following:
    - Reissue applications and reexamination proceedings are not eligible to participate in AFCP 2.0.
    - The examiner will verify that the AFCP 2.0 submission is compliant, *i.e.*, that the requirements of the program have been met (see items 1 to 7 above). For compliant submissions:
        - The examiner will review the response under 37 CFR 1.116 to determine if additional search and/or consideration (i) is necessitated by the amendment and (ii) could be completed within the time allotted under AFCP 2.0. If additional search and/or consideration is required but cannot be completed within the allotted time, the examiner will process the submission consistent with current practice concerning responses after final rejection under 37 CFR 1.116, *e.g.*, by mailing an advisory action.
        - If the examiner determines that the amendment does not necessitate additional search and/or consideration, or if the examiner determines that additional search and/or consideration is required and could be completed within the allotted time, then the examiner will consider whether the amendment places the application in condition for allowance (after completing the additional search and/or consideration, if required). If the examiner determines that the amendment does not place the application in condition for allowance, then the examiner will contact the applicant and request an interview.
            - The interview will be conducted by the examiner, and if the examiner does not have negotiation authority, a primary examiner and/or supervisory patent examiner will also participate.
            - If the applicant declines the interview, or if the interview cannot be scheduled within ten (10) calendar days from the date that the examiner first contacts the applicant, then the examiner will proceed consistent with current practice concerning responses after final rejection under 37 CFR 1.116.

Signature	Date
/Gary J. Gershik/	November 25, 2020
Name (Print/Typed) Gary J. Gershik	**Practitioner Registration No.** 39992

***Note**: This form must be signed in accordance with 37 CFR 1.33. See 37 CFR 1.4(d) for signature requirements and certifications. Submit multiple forms if more than one signature is required, see below*.*

☐ * Total of _____ forms are submitted.

Privacy Act Statement

The **Privacy Act of 1974 (P.L. 93-579)** requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (*i.e.*, GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

Electronic Acknowledgement Receipt

EFS ID:	41231107
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	25-NOV-2020
Filing Date:	07-MAR-2016
Time Stamp:	17:06:56
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1		201125_81503-AA_Amendment_DH.pdf	134251 c4f8d43d75f887fb65ccea5433f9d73cb22523b8	yes	13

	Multipart Description/PDF files in .zip description		
	Document Description	**Start**	**End**
	Applicant Arguments/Remarks Made in an Amendment	11	13
	Applicant summary of interview with examiner	9	10
	Claims	2	8
	Response After Final Action	1	1

Warnings:					
Information:					
2	After Final Consideration Program Request	201125_81503-AA_AFCP2_DH.pdf	226178 b6cbf67fdccb80060636390d206cb6ad03fa30ff	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	360429		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

REMARKS

In this amendment, applicants have amended claims 88-98, 100, and 105-107, and cancelled claim 104. The amendments to claims 88-98, 100, and 105-107 have been made in consultation with the Examiner solely to clarify the claim language.

This amendment is appropriate for entry under 37 C.F.R. § 1.116 because it amends the claims as suggested by the Examiner during the November 24, 2020 Examiner Interview. Applicants understand from the November 24, 2020 interview that the amendments to the claims made herein address all of the claim objections and indefiniteness rejections contained in the October 16, 2020 Office Action.

Finally, the enclosed amendment is eligible for consideration under the After Final Consideration Program 2.0.

Claim Objections

In the October 16, 2020 Final Office Action, the Examiner objected to claims 88, 94, 96-98, 100, and 105. The Examiner's specific objections are contained on pages 2-3 of the October 16, 2020 Final Office Action.

Applicant's response

In response, applicants have amended the claims as agreed upon during the November 24, 2020 Examiner Interview to address each of the Examiner's objections. Accordingly, the objections to claims 88, 94, 96-98, 100, and 105 should be withdrawn.

Claim Rejections – 35 U.S.C. §112

In the October 16, 2020 Final Office Action, the Examiner rejected claims 88-107 under 35 U.S.C. § 112, second paragraph, as allegedly indefinite. The Examiner's reasoning is contained on pages 3-7 of the October 16, 2020 Final Office Action.

Applicant's response

In response, applicants have amended the claims as agreed upon during the November 24, 2020 Examiner Interview to address each of the Examiner's rejections. Accordingly, this rejection to claims 88-107 should be withdrawn.

Claim Rejections – 35 U.S.C. §101

In the October 16, 2020 Final Office Action, the Examiner rejected claims 88-107 under 35 U.S.C. § 101, as allegedly directed to a judicial exception. The Examiner's reasoning is contained on pages 7-10 of the October 16, 2020 Final Office Action.

Applicant's response

In response, applicants submit that, as agreed by the Examiners during the November 17, 2020 Examiner interview, the claims are not directed to a judicial exception. Accordingly, this rejection to claims 88-107 should be withdrawn.

Conclusion

Applicants submit that based on the amendments to the claims made herein the claims are in condition for allowance. Notice to this effect is respectfully requested.

If a telephone conference would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorneys invite the Examiner to telephone them at the number provided below.

No fee is deemed necessary in connection with the filing of this Amendment. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on November 25, 2020.

/Darren Haber/

Darren Haber

Gary J. Gershik
Registration No. 39,992
Attorney for Applicants
Cooper & Dunham LLP (Customer #23432)
845 Third Avenue
6th Floor
New York, NY 10022
(212) 278-0400

SUMMARY OF NOVEMBER 17, 2020 EXAMINER INTERVIEW

On November 17, 2020 an Examiner Interview was conducted in connection with the subject application. Present at the interview were Examiner Frank W. M. Lu and Supervisory Patent Examiner (SPE) Dave Nguyen of the U.S. Patent and Trademark Office, along with applicants' representatives, Dr. Vladimir Drozdoff, Mr. Gary Gershik, and Mr. Darren Haber. Applicants acknowledge with appreciation the courtesy that Examiners Lu and Nguyen extended during the November 17, 2020 Examiner Interview.

During the November 17, 2020 Examiner Interview, the Examiners and applicants' representatives discussed the rejections of claims 88-107 under 35 U.S.C. § 112, second paragraph, for alleged indefiniteness, contained in the October 16, 2020 Office Action. The Examiners and applicants' representatives also discussed the objections to the claims contained in the October 16, 2020 Office Action. SPE Nguyen suggested that reciting "reference genome" in parts of the claims relating to mapping would be helpful in clarifying the claims. Further, SPE Nguyen acknowledged that many of the indefiniteness rejections were not adequately substantiated in the October 16, 2020 Office Action and requested that applicants' representatives schedule a further interview with Frank W. M. Lu to agree on amendments to the claims that would overcome any remaining issues with the claims.

Regarding the rejection of claims 88-107 under 35 U.S.C. § 101, SPE Nguyen agreed with applicants' representatives that the claims are not directed to a judicial exception and indicated that the rejection of claims 88-107 under 35 U.S.C. § 101 would be withdrawn.

SUMMARY OF NOVEMBER 24, 2020 EXAMINER INTERVIEW

On November 24, 2020 an Examiner Interview was conducted in connection with the subject application. Present at the interview were Examiner Frank W. M. Lu of the U.S. Patent and Trademark Office and applicants' representatives, Dr. Vladimir Drozdoff, Mr. Gary Gershik, and Mr. Darren Haber. Applicants acknowledge with appreciation the courtesy that Examiners Lu extended during the November 24, 2020 Examiner Interview.

As agreed during the November 17, 2020 Examiner Interview, during the November 24, 2020 Examiner Interview, the Examiner and applicants' representatives discussed the rejections of claims 88-107 under 35 U.S.C. § 112, second paragraph, for alleged indefiniteness and the objections to the claims, contained in the October 16, 2020 Office Action.

In particular, Examiner Lu suggested and applicants' representatives agreed to the amendments to the claims that would overcome the objections and indefiniteness rejections contained in the October 16, 2020 Office Action. Examiner Lu requested that the amendments to the claims be presented using the After Final Consideration Pilot Program 2.0 (AFCP 2.0).

The claim amendments presented herein using the AFCP 2.0 program reflect amendments to the claims suggested by the Examiner and agreed to by applicants' representatives during the November 24, 2020 Examiner Interview. Applicants understand that the amendments to the claims presented herein overcome all outstanding rejections of the claims.

AMENDMENTS TO THE CLAIMS

Please amend the claims by replacing all prior versions of the claims with the listing of claims below pursuant to 37 C.F.R. § 1.121.

Listing of Claims

1-87. (Canceled)

88. (Currently Amended) A method ~~for obtaining copy number information from genomic nucleic acids in a sample,~~ comprising:

a) obtaining segments of the genomic nucleic acids from the sample;

b) tagging the segments with nucleic acid tags, thereby generating a plurality of unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises ~~a portion corresponding to~~ a segment of the segments of the genomic nucleic acids from step (a) and ~~a portion corresponding to~~ a nucleic acid tag of the nucleic acid tags;

c) subjecting the plurality of the unique tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a reference genome by mapping ~~the subsequence of~~ each of the tag associated sequence reads of step (d) ~~corresponding to a segment of the unique tagged nucleic acid molecule~~ to a location of the reference genome; and

f) ~~at each of a plurality of locations corresponding to a segment of a unique tagged nucleic acid molecule in step (e),~~ at a plurality of locations on the reference genome, counting the number of unique tagged nucleic acid molecules that have been assigned based on the mapping of step (e) to the same location on the reference genome to obtain a count for each location of the plurality of locations on the reference genome ~~of the plurality of locations~~, thereby obtaining copy number information from genomic nucleic acids in the sample.

89. (Currently Amended) The method of claim 88, further comprising estimating a copy number [[of]]at multiple locations within a region of the ~~genome~~reference genome.

90. (Currently Amended) The method of claim 88, further comprising comparing a count from ~~the~~ counts obtained in step (f) for a location on the ~~genome~~reference genome to a count for the same location obtained from a reference sample, thereby estimating a relative copy number [[of]]at the location.

91. (Currently Amended) The method of claim 88, further comprising

g) summing ~~the counts from the~~ counts obtained in step (f) for locations on ~~the genome which comprise~~ a first region of the ~~genome~~reference genome, wherein the first region comprises more than one location;

h) summing ~~the counts from the~~ counts obtained in step (f) for locations on ~~the genome which comprise~~ a second region of the ~~genome~~reference genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the value obtained in step (g) to the value obtained in step (h),

thereby estimating the relative copy number [[of]]at the first region of the ~~genome~~reference genome to the copy number [[of]]at the second region of the ~~genome~~reference genome.

92. (Currently Amended) The method of claim 91, wherein step (h) further comprises

j) summing ~~the counts from the~~ counts obtained in step (f) for locations ~~on the genome which comprise~~ a third region of the ~~genome~~reference genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (f) for locations which comprise the second region and the sum of the counts obtained in step (j) for locations which comprise the third region.

93. (Currently Amended) The method of claim 88, further comprising (l) summing the counts from the counts obtained in step (f) for locations which comprise a region of the ~~genome~~reference genome; m) summing the counts obtained from a reference sample for the same region of the ~~genome~~reference genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative copy number [[of]]at the region of the ~~genome~~reference genome.

94. (Currently Amended) A method ~~for obtaining mRNA copy number information from mRNA transcripts in a sample,~~ comprising:

a) generating unique tagged nucleic acid molecules, comprising:

i) subjecting the mRNA transcripts in the sample to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tail of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of ~~at least one of~~ steps (i) ~~and~~ or (iii) comprise nucleic acid tags, such that each of the tagged nucleic acid molecules is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library associated with the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) corresponding to a mRNA transcript of the mRNA transcripts to a location on the cDNA library; and

e) ~~at each of a plurality of locations in step (e)~~at a plurality of locations on the cDNA library, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned based on the mapping of step (d) to the same location on the cDNA library, thereby obtaining a count for each location ~~on the cDNA library~~ of the plurality of locations on the cDNA library.

95. (Currently Amended) The method of claim 88, wherein the unique tagged nucleic acid molecules are subject to a hybrid capture method prior to PCR in step (c) or the amplified tagged nucleic acid molecules produced in step (c) are subject to hybrid capture prior to sequencing in step (d).

96. (Currently Amended) The method of claim 88, wherein each of the tagged nucleic acid molecules differs at more than one nucleotide.

97. (Currently Amended) The method of claim 88, wherein ~~tagging the segments to generate tagged nucleic acid molecules~~said tagging the segments with nucleic acid tags comprises ligation of adaptors comprising the tags to at least one end of the segments of the genomic ~~material~~nucleic acids.

98. (Currently Amended) The method of claim 88, wherein each of the tagged nucleic acid molecules comprise[[s]] more than one tag.

99. (Previously Presented) The method of claim 88, wherein the tags further comprise a sample tag.

100. (Currently Amended) The method of claim 88, wherein ~~a~~ tagged nucleic acid molecules generated from the method and that differ~~s~~ by a single nucleotide ~~is~~ are ignored ~~when counting~~during or after the counting step.

101. (Previously Presented) The method of claim 88, wherein the unique tagged nucleic acid molecules are pooled with a plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules of step (c).

102. (Previously Presented) The method of claim 101, further comprising deconvoluting the tag associated sequence reads by grouping the tag associated sequence reads.

103. (Previously Presented) The method of claim 95, wherein each of the unique tagged nucleic acid molecules subjected to hybrid capture has a nucleic acid sequence which is complementary to a capture probe on an array or on a microarray.

104. (Cancelled)

105. (Currently Amended) The method of claim 88, comprising comparing the copy number information obtained ~~for~~ from different species' genomic nucleic acids to determine the relative count of ~~those~~ the genomic nucleic acids of the different species in a population.

106. (Currently Amended) The method of claim 88, wherein the genomic nucleic acids ~~material is~~are obtained from blood or other bodily fluids.

107. (Currently Amended) The method of claim 88, wherein the genomic

nucleic acids ~~material is~~are obtained from tumor cell DNA in bodily fluid.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 12/01/2020

COOPER & DUNHAM LLP
845 Third Avenue, 6th Floor
NEW YORK, NY 10022

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

NOTIFICATION DATE	DELIVERY MODE
12/01/2020	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

DUSPTO@cooperdunham.com
patcomm-in-grp@cooperdunham.com
pdocketing@cooperdunham.com

GUARDANT HEALTH EX.1009.628

Applicant-Initiated Interview Summary	**Application No.** 15/063,278	**Applicant(s)** Hicks et al.	
	Examiner FRANK W LU	**Art Unit** 1634	**AIA (FITF) Status** No

All participants (applicant, applicants representative, PTO personnel):

(1) FRANK LU.

(2) Gary Gershik (Reg. No. 39,992).

(3) Vladimir Drozdoff, Ph.D..

(4) Darren Haber.

Date of Interview: 24 November 2020.

Type: ☑ Telephonic ☐ Video Conference
☐ Personal [copy given to: ☐ applicant ☐ applicant's representative]

Exhibit shown or demonstration conducted: ☐ Yes ☐ No.
If Yes, brief description: _____.

Issues Discussed ☐101 ☑112 ☐102 ☐103 ☐Others
(For each of the checked box(es) above, please describe below the issue and detailed description of the discussion)

Claim(s) discussed: 88-107.

Identification of prior art discussed: _____.

Substance of Interview
(For each issue discussed, provide a detailed description and indicate if agreement was reached. Some topics may include: identification or clarification of a reference or a portion thereof, claim interpretation, proposed amendments, arguments of any applied references etc...)

Applicant's representatives and examiners discussed the rejections and possible amendments. Applicant will file a response..

Applicant recordation instructions: The formal written reply to the last Office action must include the substance of the interview. (See MPEP section 713.04). If a reply to the last Office action has already been filed, applicant is given a non-extendable period of the longer of one month or thirty days from this interview date, or the mailing date of this interview summary form, whichever is later, to file a statement of the substance of the interview.

Examiner recordation instructions: Examiners must summarize the substance of any interview of record. A complete and proper recordation of the substance of an interview should include the items listed in MPEP 713.04 for complete and proper recordation including the identification of the general thrust of each argument or issue discussed, a general indication of any other pertinent matters discussed regarding patentability and the general results or outcome of the interview, to include an indication as to whether or not agreement was reached on the issues raised.

☐ Attachment

/FRANK W LU/ Primary Examiner, Art Unit 1634	November 24, 2020

Summary of Record of Interview Requirements

Manual of Patent Examining Procedure (MPEP), Section 713.04, Substance of Interview Must be Made of Record
A complete written statement as to the substance of any face-to-face, video conference, or telephone interview with regard to an application must be made of record in the application whether or not an agreement with the examiner was reached at the interview.

Title 37 Code of Federal Regulations (CFR) 1.133 Interviews
Paragraph (b)

In every instance where reconsideration is requested in view of an interview with an examiner, a complete written statement of the reasons presented at the interview as warranting favorable action must be filed by the applicant. An interview does not remove the necessity for reply to Office action as specified in §§ 1.111, 1.135. (35 U.S.C. 132)

37 CFR §1.2 Business to be transacted in writing.
All business with the Patent or Trademark Office should be transacted in writing. The personal attendance of applicants or their attorneys or agents at the Patent and Trademark Office is unnecessary. The action of the Patent and Trademark Office will be based exclusively on the written record in the Office. No attention will be paid to any alleged oral promise, stipulation, or understanding in relation to which there is disagreement or doubt.

The action of the Patent and Trademark Office cannot be based exclusively on the written record in the Office if that record is itself incomplete through the failure to record the substance of interviews.

It is the responsibility of the applicant or the attorney or agent to make the substance of an interview of record in the application file, unless the examiner indicates he or she will do so. It is the examiners responsibility to see that such a record is made and to correct material inaccuracies which bear directly on the question of patentability.

Examiners must complete an Interview Summary Form for each interview held where a matter of substance has been discussed during the interview by checking the appropriate boxes and filling in the blanks. Discussions regarding only procedural matters, directed solely to restriction requirements for which interview recordation is otherwise provided for in Section 812.01 of the Manual of Patent Examining Procedure, or pointing out typographical errors or unreadable script in Office actions or the like, are excluded from the interview recordation procedures below. Where the substance of an interview is completely recorded in an Examiners Amendment, no separate Interview Summary Record is required.

The Interview Summary Form shall be given an appropriate Paper No., placed in the right hand portion of the file, and listed on the "Contents" section of the file wrapper. In a personal interview, a duplicate of the Form is given to the applicant (or attorney or agent) at the conclusion of the interview. In the case of a telephone or video-conference interview, the copy is mailed to the applicants correspondence address either with or prior to the next official communication. If additional correspondence from the examiner is not likely before an allowance or if other circumstances dictate, the Form should be mailed promptly after the interview rather than with the next official communication.

The Form provides for recordation of the following information:
- Application Number (Series Code and Serial Number)
- Name of applicant
- Name of examiner
- Date of interview
- Type of interview (telephonic, video-conference, or personal)
- Name of participant(s) (applicant, attorney or agent, examiner, other PTO personnel, etc.)
- An indication whether or not an exhibit was shown or a demonstration conducted
- An identification of the specific prior art discussed
- An indication whether an agreement was reached and if so, a description of the general nature of the agreement (may be by attachment of a copy of amendments or claims agreed as being allowable). Note: Agreement as to allowability is tentative and does not restrict further action by the examiner to the contrary.
- The signature of the examiner who conducted the interview (if Form is not an attachment to a signed Office action)

It is desirable that the examiner orally remind the applicant of his or her obligation to record the substance of the interview of each case. It should be noted, however, that the Interview Summary Form will not normally be considered a complete and proper recordation of the interview unless it includes, or is supplemented by the applicant or the examiner to include, all of the applicable items required below concerning the substance of the interview.

A complete and proper recordation of the substance of any interview should include at least the following applicable items:
1) A brief description of the nature of any exhibit shown or any demonstration conducted,-
2) an identification of the claims discussed,
3) an identification of the specific prior art discussed,
4) an identification of the principal proposed amendments of a substantive nature discussed, unless these are already described on the Interview Summary Form completed by the Examiner,
5) a brief identification of the general thrust of the principal arguments presented to the examiner,
(The identification of arguments need not be lengthy or elaborate. A verbatim or highly detailed description of the arguments is not required. The identification of the arguments is sufficient if the general nature or thrust of the principal arguments made to the examiner can be understood in the context of the application file. Of course, the applicant may desire to emphasize and fully describe those arguments which he or she feels were or might be persuasive to the examiner.)
6) a general indication of any other pertinent matters discussed, and
7) if appropriate, the general results or outcome of the interview unless already described in the Interview Summary Form completed by the examiner.

Examiners are expected to carefully review the applicants record of the substance of an interview. If the record is not complete and accurate, the examiner will give the applicant an extendable one month time period to correct the record.

Examiner to Check for Accuracy

If the claims are allowable for other reasons of record, the examiner should send a letter setting forth the examiners version of the statement attributed to him or her. If the record is complete and accurate, the examiner should place the indication, Interview Record OK on the paper recording the substance of the interview along with the date and the examiners initials.

Doc Code: ECOMM.AUTH/ECOMM.WTDW
Doc Description: Internet Communications Authorized/Internet Communications Authorization Withdrawn

PTO/SB/439 (11-15)

AUTHORIZATION FOR INTERNET COMMUNICATIONS IN A PATENT APPLICATION OR REQUEST TO WITHDRAW AUTHORIZATION FOR INTERNET COMMUNICATIONS		
	Application No.	15/063,278
	Filing Date	March 7, 2016
	First Named Inventor	James Hicks
	Art Unit	1634
	Examiner Name	Frank Lu
	Practitioner Docket No.	81503-AA/GJG/DH

To: Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

I. To authorize permission for Internet Communications.

☑ Recognizing that Internet communications are not secure, I hereby authorize the USPTO to communicate with the undersigned and practitioners in accordance with 37 CFR 1.33 and 37 CFR 1.34 concerning any subject matter of this application via video conferencing, instant messaging, or electronic mail. I understand that a copy of these communications will be made of record in the application file. (MPEP 502.03)

II. To withdraw authorization for Internet Communications.

☐ The authorization given on _____, to the USPTO to communicate with the undersigned and any practitioner in accordance with 37 CFR 1.33 and 37 CFR 1.34 concerning any subject matter of this application via Internet communications such as video conferencing, instant messaging, or electronic mail is hereby withdrawn. I understand that the withdrawal is effective when approved rather than when received.

I am the

☐ applicant.

☑ attorney or agent of record. Registration number 39992.

☐ attorney or agent acting under 37 CFR 1.34. Registration number _____.

/Gary J. Gershik/ — Signature

December 11, 2020 — Date

Gary J. Gershik — Typed or printed name

212-278-0400 — Telephone Number

NOTE: This form must be signed in accordance with 37 CFR 1.33. See 37 CFR 1.4 for signature requirements and certifications. Juristic entities must be represented by a patent practitioner (see 37 CFR 1.31, which is applicable to any paper filed on or after September 16, 2012, that is presented on behalf of a juristic entity, regardless of application filing date). Submit multiple forms if more than one signature is required, see below*.

☐ * Total of _____ forms are submitted.

Electronic Acknowledgement Receipt

EFS ID:	41363724
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	11-DEC-2020
Filing Date:	07-MAR-2016
Time Stamp:	15:21:38
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Internet Communications Authorized	201211_81503-AA_Auth_DH.pdf	76059 a82a66c6c8fe1d6c7370f5499b93399140759056	no	1

Warnings:

GUARDANT HEALTH EX.1009.632

Information:	
Total Files Size (in bytes):	76059

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

NOTICE OF ALLOWANCE AND FEE(S) DUE

23432 7590 12/18/2020

COOPER & DUNHAM LLP
90 Park Avenue, 21st Floor
NEW YORK, NY 10016

EXAMINER
LU, FRANK WEI MIN

ART UNIT	PAPER NUMBER
1634	

DATE MAILED: 12/18/2020

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

TITLE OF INVENTION: VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
nonprovisional	SMALL	$600	$0.00	$0.00	$600	03/18/2021

THE APPLICATION IDENTIFIED ABOVE HAS BEEN EXAMINED AND IS ALLOWED FOR ISSUANCE AS A PATENT. PROSECUTION ON THE MERITS IS CLOSED. THIS NOTICE OF ALLOWANCE IS NOT A GRANT OF PATENT RIGHTS. THIS APPLICATION IS SUBJECT TO WITHDRAWAL FROM ISSUE AT THE INITIATIVE OF THE OFFICE OR UPON PETITION BY THE APPLICANT. SEE 37 CFR 1.313 AND MPEP 1308.

THE ISSUE FEE AND PUBLICATION FEE (IF REQUIRED) MUST BE PAID WITHIN THREE MONTHS FROM THE MAILING DATE OF THIS NOTICE OR THIS APPLICATION SHALL BE REGARDED AS ABANDONED. THIS STATUTORY PERIOD CANNOT BE EXTENDED. SEE 35 U.S.C. 151. THE ISSUE FEE DUE INDICATED ABOVE DOES NOT REFLECT A CREDIT FOR ANY PREVIOUSLY PAID ISSUE FEE IN THIS APPLICATION. IF AN ISSUE FEE HAS PREVIOUSLY BEEN PAID IN THIS APPLICATION (AS SHOWN ABOVE), THE RETURN OF PART B OF THIS FORM WILL BE CONSIDERED A REQUEST TO REAPPLY THE PREVIOUSLY PAID ISSUE FEE TOWARD THE ISSUE FEE NOW DUE.

HOW TO REPLY TO THIS NOTICE:

I. Review the ENTITY STATUS shown above. If the ENTITY STATUS is shown as SMALL or MICRO, verify whether entitlement to that entity status still applies.

If the ENTITY STATUS is the same as shown above, pay the TOTAL FEE(S) DUE shown above.

If the ENTITY STATUS is changed from that shown above, on PART B - FEE(S) TRANSMITTAL, complete section number 5 titled "Change in Entity Status (from status indicated above)".

For purposes of this notice, small entity fees are 1/2 the amount of undiscounted fees, and micro entity fees are 1/2 the amount of small entity fees.

II. PART B - FEE(S) TRANSMITTAL, or its equivalent, must be completed and returned to the United States Patent and Trademark Office (USPTO) with your ISSUE FEE and PUBLICATION FEE (if required). If you are charging the fee(s) to your deposit account, section "4b" of Part B - Fee(s) Transmittal should be completed and an extra copy of the form should be submitted. If an equivalent of Part B is filed, a request to reapply a previously paid issue fee must be clearly made, and delays in processing may occur due to the difficulty in recognizing the paper as an equivalent of Part B.

III. All communications regarding this application must give the application number. Please direct all communications prior to issuance to Mail Stop ISSUE FEE unless advised to the contrary.

IMPORTANT REMINDER: Maintenance fees are due in utility patents issuing on applications filed on or after Dec. 12, 1980. It is patentee's responsibility to ensure timely payment of maintenance fees when due. More information is available at www.uspto.gov/PatentMaintenanceFees.

GUARDANT HEALTH EX.1009.634

PART B - FEE(S) TRANSMITTAL

Complete and send this form, together with applicable fee(s), by mail or fax, or via EFS-Web.

By mail, send to: Mail Stop ISSUE FEE
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450

By fax, send to: (571)-273-2885

INSTRUCTIONS: This form should be used for transmitting the ISSUE FEE and PUBLICATION FEE (if required). Blocks 1 through 5 should be completed where appropriate. All further correspondence including the Patent, advance orders and notification of maintenance fees will be mailed to the current correspondence address as indicated unless corrected below or directed otherwise in Block 1, by (a) specifying a new correspondence address; and/or (b) indicating a separate "FEE ADDRESS" for maintenance fee notifications.

CURRENT CORRESPONDENCE ADDRESS (Note: Use Block 1 for any change of address)

23432 7590 12/18/2020

COOPER & DUNHAM LLP
90 Park Avenue, 21st Floor
NEW YORK, NY 10016

Note: A certificate of mailing can only be used for domestic mailings of the Fee(s) Transmittal. This certificate cannot be used for any other accompanying papers. Each additional paper, such as an assignment or formal drawing, must have its own certificate of mailing or transmission.

Certificate of Mailing or Transmission

I hereby certify that this Fee(s) Transmittal is being deposited with the United States Postal Service with sufficient postage for first class mail in an envelope addressed to the Mail Stop ISSUE FEE address above, or being transmitted to the USPTO via EFS-Web or by facsimile to (571) 273-2885, on the date below.

(Typed or printed name)
(Signature)
(Date)

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

TITLE OF INVENTION: VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
nonprovisional	SMALL	$600	$0.00	$0.00	$600	03/18/2021

EXAMINER	ART UNIT	CLASS-SUBCLASS
LU, FRANK WEI MIN	1634	435-006110

1. Change of correspondence address or indication of "Fee Address" (37 CFR 1.363).

❑ Change of correspondence address (or Change of Correspondence Address form PTO/SB/122) attached.

❑ "Fee Address" indication (or "Fee Address" Indication form PTO/SB/47; Rev 03-09 or more recent) attached. **Use of a Customer Number is required.**

2. For printing on the patent front page, list

(1) The names of up to 3 registered patent attorneys or agents OR, alternatively,

(2) The name of a single firm (having as a member a registered attorney or agent) and the names of up to 2 registered patent attorneys or agents. If no name is listed, no name will be printed.

1 _____

2 _____

3 _____

3. ASSIGNEE NAME AND RESIDENCE DATA TO BE PRINTED ON THE PATENT (print or type)

PLEASE NOTE: Unless an assignee is identified below, no assignee data will appear on the patent. If an assignee is identified below, the document must have been previously recorded, or filed for recordation, as set forth in 37 CFR 3.11 and 37 CFR 3.81(a). Completion of this form is NOT a substitute for filing an assignment.

(A) NAME OF ASSIGNEE (B) RESIDENCE: (CITY and STATE OR COUNTRY)

Please check the appropriate assignee category or categories (will not be printed on the patent) : ❑ Individual ❑ Corporation or other private group entity ❑ Government

4a. Fees submitted: ❑ Issue Fee ❑ Publication Fee (if required) ❑ Advance Order - # of Copies _____

4b. Method of Payment: *(Please first reapply any previously paid fee shown above)*

❑ Electronic Payment via EFS-Web ❑ Enclosed check ❑ Non-electronic payment by credit card (Attach form PTO-2038)

❑ The Director is hereby authorized to charge the required fee(s), any deficiency, or credit any overpayment to Deposit Account No. _____

5. **Change in Entity Status** (from status indicated above)

❑ Applicant certifying micro entity status. See 37 CFR 1.29

❑ Applicant asserting small entity status. See 37 CFR 1.27

❑ Applicant changing to regular undiscounted fee status.

NOTE: Absent a valid certification of Micro Entity Status (see forms PTO/SB/15A and 15B), issue fee payment in the micro entity amount will not be accepted at the risk of application abandonment.
NOTE: If the application was previously under micro entity status, checking this box will be taken to be a notification of loss of entitlement to micro entity status.
NOTE: Checking this box will be taken to be a notification of loss of entitlement to small or micro entity status, as applicable.

NOTE: This form must be signed in accordance with 37 CFR 1.31 and 1.33. See 37 CFR 1.4 for signature requirements and certifications.

Authorized Signature _____ Date _____

Typed or printed name _____ Registration No. _____

GUARDANT HEALTH EX 1009.635

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 12/18/2020

COOPER & DUNHAM LLP
90 Park Avenue, 21st Floor
NEW YORK, NY 10016

EXAMINER	
LU, FRANK WEI MIN	
ART UNIT	PAPER NUMBER
1634	

DATE MAILED: 12/18/2020

Determination of Patent Term Adjustment under 35 U.S.C. 154 (b)
(Applications filed on or after May 29, 2000)

The Office has discontinued providing a Patent Term Adjustment (PTA) calculation with the Notice of Allowance.

Section 1(h)(2) of the AIA Technical Corrections Act amended 35 U.S.C. 154(b)(3)(B)(i) to eliminate the requirement that the Office provide a patent term adjustment determination with the notice of allowance. See Revisions to Patent Term Adjustment, 78 Fed. Reg. 19416, 19417 (Apr. 1, 2013). Therefore, the Office is no longer providing an initial patent term adjustment determination with the notice of allowance. The Office will continue to provide a patent term adjustment determination with the Issue Notification Letter that is mailed to applicant approximately three weeks prior to the issue date of the patent, and will include the patent term adjustment on the patent. Any request for reconsideration of the patent term adjustment determination (or reinstatement of patent term adjustment) should follow the process outlined in 37 CFR 1.705.

Any questions regarding the Patent Term Extension or Adjustment determination should be directed to the Office of Patent Legal Administration at (571)-272-7702. Questions relating to issue and publication fee payments should be directed to the Customer Service Center of the Office of Patent Publication at 1-(888)-786-0101 or (571)-272-4200.

GUARDANT HEALTH EX.1009.636

Notice Requiring Inventor's Oath or Declaration	Application No. 15/063,278	Applicant(s) James Hicks	
	Examiner LU, FRANK WEI MIN	Art Unit 1634	

This notice is an attachment to the Notice of Allowability (PTOL-37), or the Notice of Allowability For A Design Application (PTOL-37D).

An inventor's oath or declaration in compliance with 37 CFR 1.63 or 1.64 executed by or with respect to each inventor has not yet been submitted.

An oath or declaration in compliance with 37 CFR 1.63, or a substitute statement in compliance with 37 CFR 1.64, executed by or with respect to each inventor (for any inventor for which a compliant oath, declaration, or substitute statement has not yet been submitted) MUST be filed no later than the date on which the issue fee is paid. See 35 U.S.C. 115(f). Failure to timely comply will result in ABANDONMENT of this application.

A properly executed inventor's oath to declaration has not been received for the following inventor(s):

If applicant previously filed one or more oaths, declarations, or substitute statements, applicant may have received an informational notice regarding deficiencies therein.

The following deficiencies are noted:

INFORMAL ACTION PROBLEMS

A new inventor's oath or declaration that identifies this application (e.g., by Application Number and filing date) is required. The inventor's oath or declaration does not comply with 37 CFR 1.63 in that it:

- does not state that the above-identified application was made or authorized to be made by the person executing the oath or declaration: **James Hicks**, **Nicholas Navin**, **Jennifer Troge**, **Zihua Wang**, and **Michael Wigler**.

Questions relating to this Notice should be directed to the Application Assistance Unit at 571-272-4200.

GUARDANT HEALTH EX.1009.637

OMB Clearance and PRA Burden Statement for PTOL-85 Part B

The Paperwork Reduction Act (PRA) of 1995 requires Federal agencies to obtain Office of Management and Budget approval before requesting most types of information from the public. When OMB approves an agency request to collect information from the public, OMB (i) provides a valid OMB Control Number and expiration date for the agency to display on the instrument that will be used to collect the information and (ii) requires the agency to inform the public about the OMB Control Number's legal significance in accordance with 5 CFR 1320.5(b).

The information collected by PTOL-85 Part B is required by 37 CFR 1.311. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 30 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, Virginia 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313-1450. Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

GUARDANT HEALTH EX.1009.638

Notice of Allowability	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address--

All claims being allowable, PROSECUTION ON THE MERITS IS (OR REMAINS) CLOSED in this application. If not included herewith (or previously mailed), a Notice of Allowance (PTOL-85) or other appropriate communication will be mailed in due course. **THIS NOTICE OF ALLOWABILITY IS NOT A GRANT OF PATENT RIGHTS.** This application is subject to withdrawal from issue at the initiative of the Office or upon petition by the applicant. See 37 CFR 1.313 and MPEP 1308.

1. ☑ This communication is responsive to the amendment filed on 11/25/2020.
   ☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.

2. ☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.

3. ☑ The allowed claim(s) is/are 88-103 and 105-107 . As a result of the allowed claim(s), you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information , please see **http://www.uspto.gov/patents/init_events/pph/index.jsp** or send an inquiry to **PPHfeedback@uspto.gov.**

4. ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

   Certified copies:

   a) ☐ All b) ☐ Some *c) ☐ None of the:

   1. ☐ Certified copies of the priority documents have been received.
   2. ☐ Certified copies of the priority documents have been received in Application No. _____ .
   3. ☐ Copies of the certified copies of the priority documents have been received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

   * Certified copies not received: _____ .

Applicant has THREE MONTHS FROM THE "MAILING DATE" of this communication to file a reply complying with the requirements noted below. Failure to timely comply will result in ABANDONMENT of this application.
THIS THREE-MONTH PERIOD IS NOT EXTENDABLE.

5. ☐ CORRECTED DRAWINGS (as "replacement sheets") must be submitted.
   ☐ including changes required by the attached Examiner's Amendment / Comment or in the Office action of Paper No./Mail Date _____ .

   Identifying indicia such as the application number (see 37 CFR 1.84(c)) should be written on the drawings in the front (not the back) of each sheet. Replacement sheet(s) should be labeled as such in the header according to 37 CFR 1.121(d).

6. ☐ DEPOSIT OF and/or INFORMATION about the deposit of BIOLOGICAL MATERIAL must be submitted. Note the attached Examiner's comment regarding REQUIREMENT FOR THE DEPOSIT OF BIOLOGICAL MATERIAL.

Attachment(s)

1. ☐ Notice of References Cited (PTO-892)
2. ☐ Information Disclosure Statements (PTO/SB/08), Paper No./Mail Date _____.
3. ☐ Examiner's Comment Regarding Requirement for Deposit of Biological Material _____.
4. ☑ Interview Summary (PTO-413), Paper No./Mail Date. 12/14/2020.
5. ☑ Examiner's Amendment/Comment
6. ☑ Examiner's Statement of Reasons for Allowance
7. ☑ Other PTOL-2323.

/FRANK W LU/ Primary Examiner, Art Unit 1634	December 14, 2020

GUARDANT HEALTH EX.1009.639

DETAILED ACTION

Reasons for Allowance

1. An examiner's amendment to the record appears below. Should the changes and/or additions be unacceptable to applicant, an amendment may be filed as provided by 37 CFR 1.312. To ensure consideration of such an amendment, it MUST be submitted no later than the payment of the issue fee.

Authorization for this examiner's amendment was given in a telephone interview with Mr. Gary Gershik (Reg. No. 39,992) on December 4, 2020.

2. The application has been amended as follows:

88. (Currently Amended) A method comprising:

a) obtaining segments of [the] genomic nucleic acids from [the] a sample;

b) tagging the segments with nucleic acid tags, thereby generating a plurality of unique tagged nucleic acid molecules, such that each of the unique tagged nucleic acid molecules comprises a segment of the segments of the genomic nucleic acids from step (a) and a nucleic acid tag of the nucleic acid tags;

c) subjecting the plurality of the unique tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

d) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (c);

e) assigning each of the amplified tagged nucleic acid molecules to a location on a reference genome from the same species from which the sample has been obtained by mapping each of the tag associated sequence reads of step (d) to a location of the reference genome; and

f) at a plurality of locations on the reference genome, counting the number of unique tagged nucleic acid molecules that have been assigned [based on the mapping of step (e)] to the same location on the reference genome based on the mapping step to obtain a count for each location of the plurality of locations on the reference genome, thereby obtaining copy number information from genomic nucleic acids in the sample.

90. (Currently Amended) The method of claim 88, further comprising comparing a count from counts obtained in step (f) for a location on the reference genome to a count for the same location on the reference genome obtained from a reference sample, thereby estimating a relative copy number at the location.

91. (Currently Amended) The method of claim 88, further comprising

g) summing counts obtained in step (f) for locations on a first region of the reference genome, wherein the first region comprises more than one location;

h) summing counts obtained in step (f) for locations on a second region of the reference genome, wherein the second region comprises a number of locations which is comparable to the number of locations of the first region;

i) comparing the [value] counts obtained in step (g) to the [value] counts obtained in step (h),

thereby estimating [the] a relative copy number at the first region of the reference genome to the copy number at the second region of the reference genome.

92. (Currently Amended) The method of claim 91, wherein step (h) further comprises

j) summing counts obtained in step (f) for locations at a third region of the reference genome, wherein the third region comprises a number of locations which is comparable to the number of locations of the first region; and

k) obtaining an average of the sum of the counts obtained in step (f) for locations [which comprise] at the second region and the sum of the counts obtained in step (j) for locations [which comprise] at the third region.

93. (Currently Amended) The method of claim 88, further comprising (l) summing the counts from the counts obtained in step (f) for locations [which comprise] at a region of the reference genome; (m) summing the counts obtained from a reference sample for the same region of the reference genome; and comparing the sums of the counts obtained in step (l) and step (m), thereby estimating a relative copy number at the region of the reference genome.

94. (Currently Amended) A method comprising:

a) generating unique tagged nucleic acid molecules, comprising:

i) subjecting [the] mRNA transcripts in [the] a sample to a polymerase reaction in the presence of primers capable of hybridizing to the polyA tails of the mRNA transcripts under conditions that promote the formation of only one complement of the mRNA transcripts, thereby generating first order derivative strands;

ii) adding a polynucleotide tail to an end of the first order derivative strands, thereby generating first order derivative strands comprising the polynucleotide tail; and

iii) subjecting the first order derivative strands comprising the polynucleotide tail to a polymerase reaction in the presence of primers capable of hybridizing to the polynucleotide tail added in step (ii) under conditions that promote the formation of only one complement of the first order derivative strands comprising the polynucleotide tail, thereby generating second order derivative strands,

wherein the primers of step (i) or (iii) comprise nucleic acid tags, such that each of the tagged nucleic acid molecules is unique, and the first order derivative strands and the second order derivative strands are the tagged nucleic acid molecules;

b) subjecting each of the tagged nucleic acid molecules to a polymerase chain reaction (PCR), thereby generating amplified tagged nucleic acid molecules;

c) generating tag associated sequence reads by sequencing the amplified tagged nucleic acid molecules of step (b);

d) assigning each of the amplified tagged nucleic acid molecules to a location on a cDNA library [associated with] produced from the mRNA transcripts by mapping the subsequence of each tag associated sequence read of said tag associated sequence reads of step (c) [corresponding to a mRNA transcript of the mRNA transcripts] to a location on the cDNA library; and

e) at a plurality of locations on the cDNA library, counting the number of the amplified tagged nucleic acid molecules having a different tag that have been assigned [based on the mapping of step (d)] to the same location on the cDNA library based on the mapping step, thereby obtaining a count for each location of the plurality of locations on the cDNA library.

95. (Currently Amended) The method of claim 88, wherein the unique tagged nucleic acid molecules are subject to a hybrid capture method prior to PCR in step (c) or the amplified tagged nucleic acid molecules produced in step (c) are subject to a hybrid capture method prior to [sequencing in] step (d).

100. (Currently Amended) The method of claim 88, wherein the tagged nucleic acid molecules generated from [the method] step (b) and that differ by a single nucleotide are ignored during or after the counting step.

101. (Currently Amended) The method of claim 88, wherein the unique tagged nucleic acid molecules are pooled with another plurality of unique tagged nucleic acid molecules having a different sample tag prior to the PCR or prior to said sequencing of the amplified tagged nucleic acid molecules of step (c).

103. (Currently Amended) The method of claim 95, wherein each of the unique tagged nucleic acid molecules subjected to the hybrid capture method has a nucleic acid sequence which is complementary to a capture probe on an array or on a microarray.

105. (Currently Amended) The method of claim 88, further comprising obtaining copy number information from genomic nucleic acids of different species in a population using the method of claim 88, and [comparing the copy number information obtained from different species' genomic nucleic acids to determine the] determining a relative count of the genomic nucleic acids of the different species in a population by comparing the copy number information obtained from the genomic nucleic acids of the different species.

107. (Currently Amended) The method of claim 88, wherein the genomic nucleic acids are obtained from tumor cell DNA in a bodily fluid.

3. The following is an examiner's statement of reasons for allowance:

Claims 88-103 and 105-107 are allowable in light of applicant's amendments filed on November 25, 2020, the terminal disclaimer filed on April 17, 2019, and the examiner's amendment. The rejections under 35 U.S.C 101 and 112 (b) have been withdrawn in view of the interview summary mailed on November 20, 2020, applicant's amendments filed on November 25, 2020, and the examiner's amendment. No prior art either alone or in combination with the other art in the record teaches/teach or reasonably suggests/suggest a method which comprises all limitations recited in claims 88 and 94.

Any comments considered necessary by applicant must be submitted no later than the payment of the issue fee and, to avoid processing delays, should preferably accompany the issue fee. Such submissions should be clearly labeled "Comments on Statement of Reasons for Allowance".

4. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center. The faxing of such papers must conform with the notices published in the Official Gazette, 1096 OG 30 (November 15, 1988), 1156 OG 61 (November 16, 1993), and 1157 OG 94 (December 28, 1993) (See 37 CAR § 1.6(d)). The CM Fax Center number is (571)273-8300.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Frank Lu, Ph.D., whose telephone number is (571)272-0746. The examiner can normally be reached on Monday-Friday from 9 A.M. to 5 P.M.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dave Nguyen, can be reached on (571)272-0731.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/FRANK W LU/
Primary Examiner, Art Unit 1634
December 14, 2020

Examiner-Initiated Interview Summary	Application No. 15/063,278	Applicant(s) Hicks et al.	
	Examiner FRANK W LU	Art Unit 1634	AIA (FITF) Status No

All participants (applicant, applicant's representative, PTO personnel):

(1) FRANK LU. (3) _____.

(2) Gary Gershik (Reg. No. 39,992). (4) _____.

Date of Interview: 14 December 2020.

Type: ☑ Telephonic ☐ Video Conference
☐ Personal [copy given to: ☐ applicant ☐ applicant's representative]

Exhibit shown or demonstration conducted: ☐ Yes ☐ No.
If Yes, brief description: _____.

Issues Discussed ☐ 101 ☐ 112 ☐ 102 ☐ 103 ☑ Others
(For each of the checked box(es) above, please describe below the issue and detailed description of the discussion)

Claim(s) discussed: 88,90-95,100-101,103,105 and 107.

Identification of prior art discussed: _____.

Substance of Interview
(For each issue discussed, provide a detailed description and indicate if agreement was reached. Some topics may include: identification or clarification of a reference or a portion thereof, claim interpretation, proposed amendments, arguments of any applied references etc...)

Mr. Gershik agreed with the examiner's amendment.

Applicant recordation instructions: It is not necessary for applicant to provide a separate record of the substance of interview.

Examiner recordation instructions: Examiners must summarize the substance of any interview of record. A complete and proper recordation of the substance of an interview should include the items listed in MPEP 713.04 for complete and proper recordation including the identification of the general thrust of each argument or issue discussed, a general indication of any other pertinent matters discussed regarding patentability and the general results or outcome of the interview, to include an indication as to whether or not agreement was reached on the issues raised.

☐ Attachment

/FRANK W LU/ Primary Examiner, Art Unit 1634	December 14, 2020

GUARDANT HEALTH EX.1009.647

Issue Classification	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	**Art Unit**
	FRANK W LU	1634

CPC							
Symbol						**Type**	**Version**
C12Q	/	1	/	6874		F	2013-01-01
G16B	/	25	/	00		I	2019-02-01
C12Q	/	1	/	6886		I	2013-01-01
C12Q	/	1	/	683		I	2013-01-01
C12Q	/	1	/	6881		I	2013-01-01
C12Q	/	2600	/	156		A	2013-01-01

CPC Combination Sets								
Symbol					**Type**	**Set**	**Ranking**	**Version**
C12Q	/	1	/	683	I	1	1	2013-01-01
C12Q	/	2525	/	155	I	1	2	2013-01-01
C12Q	/	2525	/	191	I	1	3	2013-01-01
C12Q	/	2535	/	122	I	1	4	2013-01-01
C12Q	/	2537	/	159	I	1	5	2013-01-01
C12Q	/	2537	/	16	I	1	6	2013-01-01

NONE		**Total Claims Allowed:**	
(Assistant Examiner)	(Date)	19	
/FRANK W LU/ Primary Examiner, Art Unit 1634	14 December 2020	O.G. Print Claim(s)	O.G. Print Figure
(Primary Examiner)	(Date)	1	None

U.S. Patent and Trademark Office — Part of Paper No.: 12/2020

GUARDANT HEALTH EX.1009.648

Issue Classification	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	**Art Unit**
	FRANK W LU	1634

INTERNATIONAL CLASSIFICATION				
CLAIMED				
C12P	/	19	/	34

NON-CLAIMED				
	/		/	

US ORIGINAL CLASSIFICATION	
CLASS	**SUBCLASS**
435	91.2

CROSS REFERENCES(S)						
CLASS	**SUBCLASS (ONE SUBCLASS PER BLOCK)**					

NONE		**Total Claims Allowed:**	
(Assistant Examiner)	(Date)	19	
/FRANK W LU/ Primary Examiner, Art Unit 1634	14 December 2020	O.G. Print Claim(s)	O.G. Print Figure
(Primary Examiner)	(Date)	1	None

U.S. Patent and Trademark Office

Part of Paper No.: 12/2020

GUARDANT HEALTH EX.1009.649

Issue Classification	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	**Art Unit**
	FRANK W LU	1634

☐ Claims renumbered in the same order as presented by applicant ☐ CPA ☑ T.D. ☐ R.1.47

CLAIMS

Final	Original	Final	Original	Final	Original	Final	Original	Final	Original	Final	Original	Final	Original	Final	Original
1	88	10	97	19	94										
2	89	11	98												
3	90	12	99												
4	91	13	100												
5	92	14	101												
6	93	15	102												
7	95	16	105												
8	103	17	106												
9	96	18	107												

NONE		**Total Claims Allowed:**	
(Assistant Examiner)	(Date)	19	
/FRANK W LU/ Primary Examiner, Art Unit 1634	14 December 2020	O.G. Print Claim(s)	O.G. Print Figure
(Primary Examiner)	(Date)	1	None

U.S. Patent and Trademark Office Part of Paper No.: 12/2020

GUARDANT HEALTH EX.1009.650

Search Notes	Application/Control No.	Applicant(s)/Patent Under Reexamination
	15/063,278	Hicks et al.
	Examiner	Art Unit
	FRANK W LU	1634

CPC - Searched*		
Symbol	**Date**	**Examiner**
Update	12/14/2020	/FL/

CPC Combination Sets - Searched*		
Symbol	**Date**	**Examiner**
Update	12/14/2020	/FL/

US Classification - Searched*			
Class	**Subclass**	**Date**	**Examiner**
Update		12/14/2020	/FL/

* See search history printout included with this form or the SEARCH NOTES box below to determine the scope of the search.

Search Notes		
Search Notes	**Date**	**Examiner**
Update (see attached files)	12/14/2020	/FL/

Interference Search			
US Class/CPC Symbol	**US Subclass/CPC Group**	**Date**	**Examiner**
CPC C12Q	1/683, 6874, 6881, 6886; 2600/156	12/14/2020	/FL/
CPC G16B	25/00	12/14/2020	/FL/

/FRANK LU/ Primary Examiner, Art Unit 1634	December 14, 2020

GUARDANT HEALTH EX.1009.651

AFCP 2.0 Decision	Application No.	Applicant(s)	
	15/063,278	Hicks et al.	
	Examiner	Art Unit	AIA (FITF) Status
	FRANK W LU	1634	No

This is in response to the After Final Consideration Pilot request filed 25 November 2020.

1. **Improper Request** – The AFCP 2.0 request is improper for the following reason(s) and the after final amendment submitted with the request will be treated under pre-pilot procedure.

   ☐ An AFCP 2.0 request form PTO/SB/434 (or equivalent document) was not submitted.

   ☐ A non-broadening amendment to at least one independent claim was not submitted.

   ☐ The request is not the first proper AFCP 2.0 request submitted in response to the most recent final rejection.

   ☐ Other: _____

2. **Proper Request**

   A. After final amendment submitted with the request will not be treated under AFCP 2.0.
   The after final amendment cannot be reviewed and a search conducted within the guidelines of the pilot program.

   ☐ The after final amendment will be treated under pre-pilot procedure.

   B. Updated search and/or completed additional consideration.
   The examiner performed an updated search and/or completed additional consideration of the after final amendment within the time authorized for the pilot program. The result(s) of the updated search and/or completed additional consideration are:

   ☐ 1. All of the rejections in the most recent final Office action are overcome and a Notice of Allowance is issued herewith.

   ☐ 2. The after final amendment would not overcome all of the rejections in the most recent final Office action . See attached interview summary for further details.

   ☐ 3. The after final amendment was reviewed, and it raises a new issue(s). See attached interview summary for further details.

   ☐ 4. The after final amendment raises new issues, but would overcome all of the rejections in the most recent final Office action. A decision on determining allowability could not be made within the guidelines of the pilot. See attached interview summary for further details, including any newly discovered prior art.

   ☑ 5. Other: The case has been allowed in view of the examiner's amendment.

   Examiner Note: Please attach an interview summary when necessary as described above.

GUARDANT HEALTH EX.1009.652

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s) : Cold Spring Harbor Laboratory

U.S. Serial No. : 15/063,278 Examiner : Frank W. M. Lu

Filing Date : March 7, 2016 Art Unit : 1634

Confirmation No. : 4038

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

845 Third Avenue
6th Floor
New York, NY 10022
November 25, 2020

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

OK TO ENTER: /F.W.L/

AMENDMENT UNDER 37 C.F.R. §1.116 IN RESPONSE TO OCTOBER 16, 2020 FINAL OFFICE ACTION, REQUEST FOR CONSIDERATION UNDER THE AFTER FINAL CONSIDERATION PILOT PROGRAM 2.0, AND SUMMARY OF NOVEMBER 17 AND 24, 2020 EXAMINER INTERVIEWS

This Amendment is a submission under 37 C.F.R. § 1.116 accompanying a Request for Consideration under the After Final Consideration Pilot Program 2.0 and responds to the issues raised in a October 24, 2020 Final Office Action issued in connection with the above-identified application. A response to the October 24, 2020 Final Office Action is due January 24, 2021. Accordingly, this Amendment is being timely filed.

Amendments to the Claims begin on page **2** of this paper.

A **Summary of November 17, 2020 Examiner Interview** begins on page **9** of this paper. A **Summary of November 24, 2020 Examiner Interview** begins on page **10** of this paper.

Remarks begin on page **11** of this paper.

EAST Search History

EAST Search History (Prior Art)

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	31	("nucleic acid" or cDNA or DNA or RNA or polynycleotide) same (tag or adaptor or adapter or linker) same (PCR or amplify or amplifying or amplification) same sequence near read same (genome or cDNA) same ("same" or identical) near (position or location)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	OFF	2020/12/14 23:24
L2	31	1 AND ((C12Q2535/122 OR C12Q2537/16 OR C12Q1/683 OR C12Q1/6869 OR C12Q2525/155 OR C12Q2525/191 OR C12Q2537/159 OR C12Q2563/179 OR C12Q2600/156 OR C12Q1/6874 OR C12Q1/6881 OR C12Q1/6886 OR C12Q1/6895 OR C12Q2600/13 OR C12Q2600/154 OR C12Q2600/158 OR G16B25/00 OR A01H1/04 OR C12N15/113 OR C12N15/8218 OR C12N15/8241 OR C12N15/8261).CPC.)	US-PGPUB; USPAT; USOCR; FPRS; EPO; JPO; DERWENT; IBM_TDB	OR	OFF	2020/12/14 23:25

12/14/2020 11:46:11 PM
C:\Users\flu\Documents\EAST\Workspaces\~auto.02042018.005450.wsp

```
(FILE 'HOME' ENTERED AT 23:36:23 ON 14 DEC 2020)

     FILE 'MEDLINE, CAPLUS, BIOSIS, SCISEARCH, EMBASE' ENTERED AT 23:37:01
ON
     14 DEC 2020
L1     13743934 S NUCLEIC ACID OR CDNA OR DNA OR RNA OR POLYNYCLEOTIDE
L2       867655 S TAG OR ADAPTOR OR ADAPTER OR LINKER
L3      4061583 S PCR OR AMPLIFY OR AMPLIFYING OR AMPLIFICATION
L4        14654 S  SEQUENCE READ
L5      3586799 S GENOME OR CDNA
L6        56317 S "SAME" POSITION OR "SAME" LOCATION
L7         4385 S IDENTICAL  POSITION OR IDENTICAL LOCATION
L8            3 S L1 AND L2 AND L3 AND L4 AND L5 AND L6
L9            0 S L1 AND L2 AND L3 AND L4 AND L5 AND L7
L10         206 S L1 AND L2 AND L3 AND L4 AND L5
```

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	:	Cold Spring Harbor Laboratory
Inventors	:	James Hicks et al.
U.S. Appl. No.	:	15/063,278
Filed	:	March 7, 2016
Confirmation No.	:	4038
For	:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

90 Park Avenue, 21st Floor
New York, New York 10016
January 21, 2021

BY EFS
Commissioner for Patents
Alexandria, VA

COMMUNICATION FORWARDING CORRECTED APPLICATION DATA SHEET

Applicant submits herewith a corrected Application Data Sheet (ADS) in compliance with 37 CFR 1.76(c). The information being added is indicated by underlining and information being removed has been stricken-through. This corrected ADS updates the mailing addresses of the five inventors, updates the email address under "Correspondence Information," and updates the Prior Application Status of U.S. Application No. 13/278,333 under "Domestic Benefit/National Stage Information."

No fee is deemed necessary in connection with the filing of this Communication. However, if any fee is required authorization is hereby given to charge the amount of any such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorney for Applicant
Cooper & Dunham LLP
90 Park Avenue
21st Floor
New York, New York 10016
(212) 278-0400

Certificate of Transmission
I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on January 21, 2021.

/Jessica G. Oshanani/
Jessica G. Oshanani

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

The application data sheet is part of the provisional or nonprovisional application for which it is being submitted. The following form contains the bibliographic data arranged in a format specified by the United States Patent and Trademark Office as outlined in 37 CFR 1.76.
This document may be completed electronically and submitted to the Office in electronic format using the Electronic Filing System (EFS) or the document may be printed and included in a paper filed application.

Secrecy Order 37 CFR 5.2:

☐ Portions or all of the application associated with this Application Data Sheet may fall under a Secrecy Order pursuant to 37 CFR 5.2 (Paper filers only. Applications that fall under Secrecy Order may not be filed electronically.)

Inventor Information:

Inventor 1 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	James		Hicks	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	~~Locust Valley~~ Palm Desert	State/Province	~~NY~~ CA	Country of Residence	US

Mailing Address of Inventor:

Address 1	~~3 Great Meadows Road~~ 380 Gold Canyon Drive		
Address 2			
City	~~Locust Valley~~ Palm Desert	State/Province	~~NY~~ CA
Postal Code	~~11560~~ 92211	Country	US

Inventor 2 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Nicholas		Navin	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	~~Houston~~ Bellaire	State/Province	TX	Country of Residence	US

Mailing Address of Inventor:

Address 1	~~1621 Wichita Street~~ 4507 Holly Street		
Address 2			
City	~~Houston~~ Bellaire	State/Province	TX
Postal Code	~~77004~~ 77401	Country	US

Inventor 3 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Jennifer		~~Trege~~ Clarke	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

City	~~Oyster Bay~~ Exeter	State/Province	~~NY~~ NH	Country of Residence	US

Mailing Address of Inventor:

Address 1	~~100 Simcoe Street~~ 61 Acadia Lane #402		
Address 2			
City	~~Oyster Bay~~ Exeter	State/Province	~~NY~~ NH
Postal Code	~~11771~~ 03833	Country	US

Inventor 4 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Zihua		Wang	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	~~Huntington~~ East Northport	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	~~13 Meadowlawn Street~~ 182 Ketay Drive South		
Address 2			
City	~~Huntington~~ East Northport	State/Province	NY
Postal Code	~~11743~~ 11731	Country	US

Inventor 5 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Michael		Wigler	

Residence Information (Select One) (•) US Residency ( ) Non US Residency ( ) Active US Military Service

City	Cold Spring Harbor	State/Province	NY	Country of Residence	US

Mailing Address of Inventor:

Address 1	~~52 Snake Hill Road~~ 40 Walnut Tree Lane		
Address 2			
City	Cold Spring Harbor	State/Province	NY
Postal Code	11724	Country	US

All Inventors Must Be Listed - Additional Inventor Information blocks may be generated within this form by selecting the **Add** button. Add

Correspondence Information:

Enter either Customer Number or complete the Correspondence Information section below.
For further information see 37 CFR 1.33(a).

☐ **An Address is being provided for the correspondence Information of this application.**

GUARDANT HEALTH EX.1009.658

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Customer Number	23432		
Email Address	~~patentdocketing@cooperdunham.com~~ pdocketing@cooperdunham.com	Add Email	Remove Email

Application Information:

Title of the Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION		
Attorney Docket Number	81503-AA/GJG/AWG/AC	**Small Entity Status Claimed**	☒
Application Type	Nonprovisional		
Subject Matter	Utility		
Total Number of Drawing Sheets (if any)	15	**Suggested Figure for Publication (if any)**	

Filing By Reference:

Only complete this section when filing an application by reference under 35 U.S.C. 111(c) and 37 CFR 1.57(a). Do not complete this section if application papers including a specification and any drawings are being filed. Any domestic benefit or foreign priority information must be provided in the appropriate section(s) below (i.e., "Domestic Benefit/National Stage Information" and "Foreign Priority Information").

For the purposes of a filing date under 37 CFR 1.53(b), the description and any drawings of the present application are replaced by this reference to the previously filed application, subject to conditions and requirements of 37 CFR 1.57(a).

Application number of the previously filed application	Filing date (YYYY-MM-DD)	Intellectual Property Authority or Country

Publication Information:

☐ Request Early Publication (Fee required at time of Request 37 CFR 1.219)

☐ **Request Not to Publish.** I hereby request that the attached application not be published under 35 U.S.C. 122(b) and certify that the invention disclosed in the attached application **has not and will not** be the subject of an application filed in another country, or under a multilateral international agreement, that requires publication at eighteen months after filing.

Representative Information:

Representative information should be provided for all practitioners having a power of attorney in the application. Providing this information in the Application Data Sheet does not constitute a power of attorney in the application (see 37 CFR 1.32).
Either enter Customer Number or complete the Representative Name section below. If both sections are completed the customer Number will be used for the Representative Information during processing.

Please Select One:	◉ Customer Number	○ US Patent Practitioner	○ Limited Recognition (37 CFR 11.9)
Customer Number	23432		

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Domestic Benefit/National Stage Information:

This section allows for the applicant to either claim benefit under 35 U.S.C. 119(e), 120, 121, 365(c), or 386(c) or indicate National Stage entry from a PCT application. Providing benefit claim information in the Application Data Sheet constitutes the specific reference required by 35 U.S.C. 119(e) or 120, and 37 CFR 1.78.
When referring to the current application, please leave the "Application Number" field blank.

Prior Application Status	~~Pending~~ Patented		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
	Continuation of	13/278333	2011-10-21
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/510579	2011-07-22
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
13/278333	Claims benefit of provisional	61/406067	2010-10-22

Additional Domestic Benefit/National Stage Data may be generated within this form by selecting the **Add** button.

Foreign Priority Information:

This section allows for the applicant to claim priority to a foreign application. Providing this information in the application data sheet constitutes the claim for priority as required by 35 U.S.C. 119(b) and 37 CFR 1.55. When priority is claimed to a foreign application that is eligible for retrieval under the priority document exchange program (PDX)[i] the information will be used by the Office to automatically attempt retrieval pursuant to 37 CFR 1.55(i)(1) and (2). Under the PDX program, applicant bears the ultimate responsibility for ensuring that a copy of the foreign application is received by the Office from the participating foreign intellectual property office, or a certified copy of the foreign priority application is filed, within the time period specified in 37 CFR 1.55(g)(1).

			Remove
Application Number	Country[i]	Filing Date (YYYY-MM-DD)	Access Code[i] (if applicable)

Additional Foreign Priority Data may be generated within this form by selecting the **Add** button.

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

☐ This application (1) claims priority to or the benefit of an application filed before March 16, 2013 and (2) also contains, or contained at any time, a claim to a claimed invention that has an effective filing date on or after March 16, 2013.
NOTE: By providing this statement under 37 CFR 1.55 or 1.78, this application, with a filing date on or after March 16, 2013, will be examined under the first inventor to file provisions of the AIA.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Authorization or Opt-Out of Authorization to Permit Access:

When this Application Data Sheet is properly signed and filed with the application, applicant has provided written authority to permit a participating foreign intellectual property (IP) office access to the instant application-as-filed (see paragraph A in subsection 1 below) and the European Patent Office (EPO) access to any search results from the instant application (see paragraph B in subsection 1 below).

Should applicant choose not to provide an authorization identified in subsection 1 below, applicant **must opt-out** of the authorization by checking the corresponding box A or B or both in subsection 2 below.

NOTE: This section of the Application Data Sheet is **ONLY** reviewed and processed with the **INITIAL** filing of an application. After the initial filing of an application, an Application Data Sheet cannot be used to provide or rescind authorization for access by a foreign IP office(s). Instead, Form PTO/SB/39 or PTO/SB/69 must be used as appropriate.

1. Authorization to Permit Access by a Foreign Intellectual Property Office(s)

A. Priority Document Exchange (PDX) - Unless box A in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the European Patent Office (EPO), the Japan Patent Office (JPO), the Korean Intellectual Property Office (KIPO), the State Intellectual Property Office of the People's Republic of China (SIPO), the World Intellectual Property Organization (WIPO), and any other foreign intellectual property office participating with the USPTO in a bilateral or multilateral priority document exchange agreement in which a foreign application claiming priority to the instant patent application is filed, access to: (1) the instant patent application-as-filed and its related bibliographic data, (2) any foreign or domestic application to which priority or benefit is claimed by the instant application and its related bibliographic data, and (3) the date of filing of this Authorization. See 37 CFR 1.14(h)(1).

B. Search Results from U.S. Application to EPO - Unless box B in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the EPO access to the bibliographic data and search results from the instant patent application when a European patent application claiming priority to the instant patent application is filed. See 37 CFR 1.14(h)(2).

The applicant is reminded that the EPO's Rule 141(1) EPC (European Patent Convention) requires applicants to submit a copy of search results from the instant application without delay in a European patent application that claims priority to the instant application.

2. Opt-Out of Authorizations to Permit Access by a Foreign Intellectual Property Office(s)

☐ A. Applicant **DOES NOT** authorize the USPTO to permit a participating foreign IP office access to the instant application-as-filed. If this box is checked, the USPTO will not be providing a participating foreign IP office with any documents and information identified in subsection 1A above.

☐ B. Applicant **DOES NOT** authorize the USPTO to transmit to the EPO any search results from the instant patent application. If this box is checked, the USPTO will not be providing the EPO with search results from the instant application.

NOTE: Once the application has published or is otherwise publicly available, the USPTO may provide access to the application in accordance with 37 CFR 1.14.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Applicant Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

Applicant 1

If the applicant is the inventor (or the remaining joint inventor or inventors under 37 CFR 1.45), this section should not be completed. The information to be provided in this section is the name and address of the legal representative who is the applicant under 37 CFR 1.43; or the name and address of the assignee, person to whom the inventor is under an obligation to assign the invention, or person who otherwise shows sufficient proprietary interest in the matter who is the applicant under 37 CFR 1.46. If the applicant is an applicant under 37 CFR 1.46 (assignee, person to whom the inventor is obligated to assign, or person who otherwise shows sufficient proprietary interest) together with one or more joint inventors, then the joint inventor or inventors who are also the applicant should be identified in this section.

Clear

◯ Assignee	◯ Legal Representative under 35 U.S.C. 117	◯ Joint Inventor
◯ Person to whom the inventor is obligated to assign.	◯ Person who shows sufficient proprietary interest	

If applicant is the legal representative, indicate the authority to file the patent application, the inventor is:

Name of the Deceased or Legally Incapacitated Inventor:

If the Applicant is an Organization check here. ☒

Organization Name	Cold Spring Harbor Laboratory

Mailing Address Information For Applicant:

Address 1	One Bungtown Road		
Address 2			
City	Cold Spring Harbor	**State/Province**	NY
Country	US	Postal Code	11724
Phone Number		Fax Number	
Email Address			

Additional Applicant Data may be generated within this form by selecting the Add button.

Assignee Information including Non-Applicant Assignee Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

Assignee 1

Complete this section if assignee information, including non-applicant assignee information, is desired to be included on the patent application publication. An assignee-applicant identified in the "Applicant Information" section will appear on the patent application publication as an applicant. For an assignee-applicant, complete this section only if identification as an assignee is also desired on the patent application publication.

If the Assignee or Non-Applicant Assignee is an Organization check here. ☒

Organization Name	Cold Spring Harbor Laboratory

Mailing Address Information For Assignee including Non-Applicant Assignee:

Address 1	One Bungtown Road		
Address 2			
City	Cold Spring Harbor	**State/Province**	NY
Country	US	Postal Code	11724
Phone Number		Fax Number	
Email Address			

Additional Assignee or Non-Applicant Assignee Data may be generated within this form by selecting the Add button.

Signature:

NOTE: This Application Data Sheet must be signed in accordance with 37 CFR 1.33(b). **However, if this Application Data Sheet is submitted with the INITIAL filing of the application and either box A or B is not checked in subsection 2 of the "Authorization or Opt-Out of Authorization to Permit Access" section, then this form must also be signed in accordance with 37 CFR 1.14(c).**

This Application Data Sheet **must** be signed by a patent practitioner if one or more of the applicants is a **juristic entity** (e.g., corporation or association). If the applicant is two or more joint inventors, this form must be signed by a patent practitioner, **all** joint inventors who are the applicant, or one or more joint inventor-applicants who have been given power of attorney (e.g., see USPTO Form PTO/AIA/81) on behalf of **all** joint inventor-applicants.

See 37 CFR 1.4(d) for the manner of making signatures and certifications.

Signature	/Gary J. Gershik/			Date (YYYY-MM-DD)	2021-01-21 ~~2019-04-17~~
First Name	Gary	Last Name	Gershik	Registration Number	39992

Additional Signature may be generated within this form by selecting the Add button.

PTO/AIA/14 (11-15)
Approved for use through 04/30/2017. OMB 0651-0032
U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

Application Data Sheet 37 CFR 1.76	Attorney Docket Number	81503-AA/GJG/AWG/AC
	Application Number	
Title of Invention	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION	

This collection of information is required by 37 CFR 1.76. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 23 minutes to complete, including gathering, preparing, and submitting the completed application data sheet form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1 The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.

2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.

3 A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.

4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).

5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent CooperationTreaty.

6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).

7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.

8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.

9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

GUARDANT HEALTH EX.1009.666

Electronic Acknowledgement Receipt

EFS ID:	41710309
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	21-JAN-2021
Filing Date:	07-MAR-2016
Time Stamp:	17:14:51
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Transmittal Letter	210121_81503-AA_Comm_Fwd_Corrected_ADS_JGO.pdf	75992 4a4152f49b5a3abf5b85ba29f50e2bbfa4dff9d9	no	1

Warnings:

GUARDANT HEALTH EX.1009.667

Information:					
2	Application Data Sheet	210121_81503-AA_ADS_JGO.pdf	148100 33f58c02b1f4819152a807d8b2a91b23de01a362	no	10
Warnings:					
Information:					
This is not an USPTO supplied ADS fillable form					
			Total Files Size (in bytes):	224092	

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

Docket No. 81503-AA/GJG/AWG/AC/JGO

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Cold Spring Harbor Laboratory

Inventors: James Hicks et al.

Serial No. : 15/063,278 Examiner: Frank Wei Min Lu

Filed : March 7, 2016 Group Art Unit: 1634

For : VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Conf. No. : 4038

90 Park Avenue – 21st Floor
New York, NY 10016
January 21, 2021

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

COMMUNICATION SUBMITTING DECLARATIONS COMPLYING WITH 37 CFR §1.63 IN RESPONSE TO DECEMBER 18, 2020 NOTICE REQUIRING INVENTOR'S OATH OR DECLARATION

This Communication is submitted in reply to the Notice Requiring Inventor's Oath or Declaration mailed December 18, 2020 in connection with the above-identified application. The Notice, a copy of which is attached hereto as **Exhibit A**, indicates that an inventor's oath or declaration is required because an inventor's oath or declaration in compliance with 37 CFR 1.63 or 1.64 has not yet been submitted. The Notice further indicates that a signed oath or declaration must be filed no later than the date on which the issue fee is paid. The issue fee is due March 18, 2021 and has not yet been paid. Accordingly, this Communication and the attached Declaration are being timely filed.

Specifically, applicants submit as **Exhibit 1** attached hereto, a copy of a Declaration signed by inventor James Hicks. Applicant also submits a copy of a Declaration signed by inventor Nicholas Navin as **Exhibit 2** attached hereto. Applicant also submits a copy of a Declaration signed by inventor Jennifer Clarke as **Exhibit 3** attached hereto. Applicant also submits a copy of a Declaration signed by inventors Zihua Wang and Michael Wigler as **Exhibit 4** attached hereto. In compliance with 37 C.F.R. §1.63, each signed Declaration refers to the application's above-

identified U.S. serial number and filing date and states that the application was made or was authorized to be made by the inventor.

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicants' undersigned attorney invites the Examiner to telephone him at the number provided below.

No fee is deemed necessary in connection with the filing of this Communication. However, if any fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik
Registration No. 39,992
Attorneys for Applicants
Cooper & Dunham LLP
90 Park Avenue – 21st Floor
New York, NY 10016
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on January 21, 2021.

/Jessica G. Oshanani/
Jessica G. Oshanani

Notice Requiring Inventor's Oath or Declaration	Application No. 15/063,278	Applicant(s) James Hicks	
	Examiner LU, FRANK WEI MIN	Art Unit 1634	

This notice is an attachment to the Notice of Allowability (PTOL-37), or the Notice of Allowability For A Design Application (PTOL-37D).

An inventor's oath or declaration in compliance with 37 CFR 1.63 or 1.64 executed by or with respect to each inventor has not yet been submitted.

An oath or declaration in compliance with 37 CFR 1.63, or a substitute statement in compliance with 37 CFR 1.64, executed by or with respect to each inventor (for any inventor for which a compliant oath, declaration, or substitute statement has not yet been submitted) MUST be filed no later than the date on which the issue fee is paid. See 35 U.S.C. 115(f). Failure to timely comply will result in ABANDONMENT of this application.

A properly executed inventor's oath to declaration has not been received for the following inventor(s):

If applicant previously filed one or more oaths, declarations, or substitute statements, applicant may have received an informational notice regarding deficiencies therein.

The following deficiencies are noted:

INFORMAL ACTION PROBLEMS

A new inventor's oath or declaration that identifies this application (e.g., by Application Number and filing date) is required. The inventor's oath or declaration does not comply with 37 CFR 1.63 in that it:

- does not state that the above-identified application was made or authorized to be made by the person executing the oath or declaration: **James Hicks**, **Nicholas Navin**, **Jennifer Troge**, **Zihua Wang**, and **Michael Wigler**.

Questions relating to this Notice should be directed to the Application Assistance Unit at 571-272-4200.

Applicant(s): Cold Spring Harbor Laboratory
U.S. Serial No.: 15/063,278
Filed: March 7, 2016
Exhibit A

GUARDANT HEALTH EX.1009.671

OMB Clearance and PRA Burden Statement for PTOL-85 Part B

The Paperwork Reduction Act (PRA) of 1995 requires Federal agencies to obtain Office of Management and Budget approval before requesting most types of information from the public. When OMB approves an agency request to collect information from the public, OMB (i) provides a valid OMB Control Number and expiration date for the agency to display on the instrument that will be used to collect the information and (ii) requires the agency to inform the public about the OMB Control Number's legal significance in accordance with 5 CFR 1320.5(b).

The information collected by PTOL-85 Part B is required by 37 CFR 1.311. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 30 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, Virginia 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313-1450. Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

GUARDANT HEALTH EX.1009.672

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which: (check one)

_____ *is attached hereto.*

__X__ *was filed on* __March 7, 2016__ *as*

U.S. Application No __15/063,278__

and was amended __March 7, 2016__
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above. The application is made or is authorized to be made, or was made or was authorized to be made by me.

I am aware of the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Expired
61/406,067	October 22, 2010	Expired

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
13/278,333	October 21, 2011	Patented

And I hereby appoint

John P. White (Reg. No. 28,678); Paul Teng (Reg. No. 40,837); and Gary J. Gershik (Reg. No. 39,992).

and each of them, all c/o Cooper & Dunham LLP, 90 Park Avenue, 21st Floor, New York, New York 10016, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to:

Gary J. Gershik *Reg.No.* 39,992
Cooper & Dunham, LLP (Customer Number 23432)
90 Park Avenue
21st Floor
New York, New York 10016
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment of not more than five (5) years, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor	James Hicks
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	3 Great Meadow Road, Locust Valley, NY 11560
Post Office Address	See residence address

Full name of additional joint inventor (if any)	Nicholas Navin
Inventor's signature	*Date of signature* 1/12/2021
Citizenship	United States
Residence	4507 Holly St., Bellaire, TX 77401
Post Office Address	See residence address

Full name of additional joint inventor (if any)	Jennifer Troge
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	100 Simcoe St., Oyster Bay, NY 11771
Post Office Address	See residence address

Full name of additional joint inventor (if any) Zihua Wang

Inventor's signature _____ *Date of signature* _____

Citizenship China

Residence 13 Meadowlawn St., Huntington, NY 11743

Post Office Address See residence address

Full name of additional joint inventor (if any) Michael Wigler

Inventor's signature _____ *Date of signature* _____

Citizenship United States

Residence 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Post Office Address See residence address

Docket Number: 81503-AA/GJG/AWG/JGO

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which: (check one)

_____ *is attached hereto.*

__X__ *was filed on* __March 7, 2016__ *as*

U.S. Application No __15/063,278__

and was amended __March 7, 2016__
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above. The application is made or is authorized to be made, or was made or was authorized to be made by me.

I am aware of the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Expired
61/406,067	October 22, 2010	Expired

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
13/278,333	October 21, 2011	Patented

And I hereby appoint

John P. White (Reg. No. 28,678); Paul Teng (Reg. No. 40,837); and Gary J. Gershik (Reg. No. 39,992).

and each of them, all c/o Cooper & Dunham LLP, 90 Park Avenue, 21st Floor, New York, New York 10016, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to:

Gary L. Gerstein Reg No. 39,992
Cooper & Dunham, LLP (Customer Number 23432)
30 Park Avenue
21st Floor
New York, New York 10016
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment of not more than five (5) years, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor: James Hicks
Inventor's signature: _____ Date of signature: _____
Citizenship: United States
Residence: 3 Great Meadow Road, Locust Valley, NY 11560
Post Office Address: See residence address

Full name of additional joint inventor (if any): Nicholas Navin
Inventor's signature: _____ Date of signature: _____
Citizenship: United States
Residence: 4507 Holly St., Bellaire, TX 77401
Post Office Address: See residence address

Full name of additional joint inventor (if any): Jennifer Clarke
Inventor's signature: _____ Date of signature: 01-21-2021
Citizenship: United States
Residence: 61 Acadia Lane #402, Exeter, NH 03833
Post Office Address: See residence address

Full name of additional joint inventor (if any) Zihua Wang

Inventor's signature _____ *Date of signature* _____

Citizenship China

Residence 13 Meadowlawn St., Huntington, NY 11743

Post Office Address See residence address

Full name of additional joint inventor (if any) Michael Wigler

Inventor's signature _____ *Date of signature* _____

Citizenship United States

Residence 52 Snake Hill Road, Cold Spring Harbor, NY 11724

Post Office Address See residence address

Docket Number: 81503-AA/GJG/AWG/JGO

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which: (check one)

_____ *is attached hereto.*

__X__ *was filed on* March 7, 2016 *as*

U.S. Application No 15/063,278

and was amended March 7, 2016
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above. The application is made or is authorized to be made, or was made or was authorized to be made by me.

I am aware of the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

GUARDANT HEALTH EX.1009.681

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Expired
61/406,067	October 22, 2010	Expired

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
13/278,333	October 21, 2011	Patented

And I hereby appoint

John P. White (Reg. No. 28,678); Paul Teng (Reg. No. 40,837); and Gary J. Gershik (Reg. No. 39,992).

and each of them, all c/o Cooper & Dunham LLP, 90 Park Avenue, 21st Floor, New York, New York 10016, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to:

Gary J. Gershik *Reg.No.* 39,992
Cooper & Dunham, LLP (Customer Number 23432)
90 Park Avenue
21st Floor
New York, New York 10016
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment of not more than five (5) years, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor	James Hicks
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	3 Great Meadow Road, Locust Valley, NY 11560
Post Office Address	See residence address

Full name of additional joint inventor (if any)	Nicholas Navin
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	1621 Wichita St., Houston, TX 77004
Post Office Address	See residence address

Full name of additional joint inventor (if any)	Jennifer Troge
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	100 Simcoe St., Oyster Bay, NY 11771
Post Office Address	See residence address

Full name of additional joint inventor (if any) Zihua Wang

Inventor's signature [signature] *Date of signature* 1/11/21

Citizenship China

Residence ~~13 Meadowlawn St., Huntington, NY 11743~~

Post Office Address See residence address

182 Ketay Dr S., East Northport, NY 11731 (ZW)

Full name of additional joint inventor (if any) Michael Wigler

Inventor's signature [signature] *Date of signature* 1/11/21

Citizenship United States

Residence ~~32 Snake Hill Road, Cold Spring Harbor, NY 11724~~

Post Office Address See residence address

40 Walnut Tree Lane
Cold Spring Harbor NY
11724 (MW)

Docket Number: 81503-AA/GJG/AWG/JGO

DECLARATION AND POWER OF ATTORNEY

As a below-named inventor, I hereby declare that:

My residence, post office address, and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

the specification of which: (check one)

_____ *is attached hereto.*

__X__ *was filed on* __March 7, 2016__ *as*

U.S. Application No __15/063,278__

and was amended __March 7, 2016__
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above. The application is made or is authorized to be made, or was made or was authorized to be made by me.

I am aware of the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, Section 119(a)-(d) or Section 365(b) of any foreign application(s) for patent or inventor's certificate, or Section 365(a) of any PCT International Application which designated at least one country other than the United States, listed below. I have also identified below any foreign application for patent or inventor's certificate, or PCT International Application having a filing date before that of the earliest application from which priority is claimed:

Prior Foreign Application(s)			*Priority Claimed*	
Number	*Country*	*Filing Date*	*Yes*	*No*
N/A				

GUARDANT HEALTH EX.1009.685

I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below:

Provisional Application No.	*Filing Date*	*Status*
61/510,579	July 22, 2011	Expired
61/406,067	October 22, 2010	Expired

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States Application(s), or Section 365(c) of any PCT International Application(s) designating the United States listed below. Insofar as this application discloses and claims subject matter in addition to that disclosed in any such prior Application in the manner provided by the first paragraph of Title 35, United States Code, Section 112, I acknowledge the duty to disclose to the United States Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56, which became available between the filing date(s) of such prior Application(s) and the national or PCT international filing date of this application:

Application Serial No.	*Filing Date*	*Status*
13/278,333	October 21, 2011	Patented

And I hereby appoint

John P. White (Reg. No. 28,678); Paul Teng (Reg. No. 40,837); and Gary J. Gershik (Reg. No. 39,992).

and each of them, all c/o Cooper & Dunham LLP, 90 Park Avenue, 21st Floor, New York, New York 10016, my attorneys, each with full power of substitution and revocation, to prosecute this application, to make alterations and amendments therein, to receive the patent, to transact all business in the Patent and Trademark Office connected therewith and to file any International Applications which are based thereon under the provisions of the Patent Cooperation Treaty.

Please address all communications, and direct all telephone calls, regarding this application to:

Gary J. Gershik *Reg.No.* 39,992
Cooper & Dunham, LLP (Customer Number 23432)
90 Park Avenue
21st Floor
New York, New York 10016
Tel. (212) 278-0400

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment of not more than five (5) years, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Full name of sole or first joint inventor James Hicks
Inventor's signature *Date of signature* JAN 11, 2021
Citizenship United States
Residence ~~3 Great Meadow Road, Locust Valley, NY 11560~~
Post Office Address See residence address 380 Gold Canyon Drive Palm Desert CA 92211

Full name of additional joint inventor (if any) Nicholas Navin
Inventor's signature *Date of signature*
Citizenship United States
Residence 1621 Wichita St., Houston, TX 77004
Post Office Address See residence address

Full name of additional joint inventor (if any) Jennifer Troge
Inventor's signature *Date of signature*
Citizenship United States
Residence 100 Simcoe St., Oyster Bay, NY 11771
Post Office Address See residence address

Full name of additional joint inventor (if any)	Zihua Wang
Inventor's signature	*Date of signature*
Citizenship	China
Residence	13 Meadowlawn St., Huntington, NY 11743
Post Office Address	See residence address

Full name of additional joint inventor (if any)	Michael Wigler
Inventor's signature	*Date of signature*
Citizenship	United States
Residence	52 Snake Hill Road, Cold Spring Harbor, NY 11724
Post Office Address	See residence address

Electronic Acknowledgement Receipt

EFS ID:	41710409
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	21-JAN-2021
Filing Date:	07-MAR-2016
Time Stamp:	17:20:29
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	no

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Transmittal Letter	210121_81503-AA_Comm_Fwd_Dec_JGO.pdf	68447 de196769b3289cc1aacd55a88dca5ffe46932701	no	2

Warnings:

GUARDANT HEALTH EX.1009.689

Information:					
2	Applicant Arguments/Remarks Made in an Amendment	210121_81503-AA_Exhibit_A_JGO.pdf	1202509 d97494162e71507fb021ac8a06ead932d8c7b070	no	2
Warnings:					
Information:					
3	Oath or Declaration filed	210121_81503-AA_Exhibit_2_JGO.pdf	309411 8dec10e52665750afc24d33884c49ac0794eb73c	no	4
Warnings:					
Information:					
4	Oath or Declaration filed	210121_81503-AA_Exhibit_3_JGO.pdf	263972 3caebe210adec99b2a7826a8893c80af8aa2e95d	no	4
Warnings:					
Information:					
5	Oath or Declaration filed	210121_81503-AA_Exhibit_4_JGO.pdf	1253364 36a85203c3e5a45fc0b25add042370fcb9f7b591	no	4
Warnings:					
Information:					
6	Oath or Declaration filed	210121_81503-AA_Exhibit_1_JGO.pdf	6463374 b946496637445b034693962f5692bf5d4386f156	no	4
Warnings:					
Information:					
		Total Files Size (in bytes):	9561077		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

PART B - FEE(S) TRANSMITTAL

Complete and send this form, together with applicable fee(s), by mail or fax, or via EFS-Web.

By mail, send to: Mail Stop ISSUE FEE
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450

By fax, send to: (571)-273-2885

INSTRUCTIONS: This form should be used for transmitting the ISSUE FEE and PUBLICATION FEE (if required). Blocks 1 through 5 should be completed where appropriate. All further correspondence including the Patent, advance orders and notification of maintenance fees will be mailed to the current correspondence address as indicated unless corrected below or directed otherwise in Block 1, by (a) specifying a new correspondence address; and/or (b) indicating a separate "FEE ADDRESS" for maintenance fee notifications.

CURRENT CORRESPONDENCE ADDRESS (Note: Use Block 1 for any change of address)

23432 7590 12/18/2020

COOPER & DUNHAM LLP
90 Park Avenue, 21st Floor
NEW YORK, NY 10016

Note: A certificate of mailing can only be used for domestic mailings of the Fee(s) Transmittal. This certificate cannot be used for any other accompanying papers. Each additional paper, such as an assignment or formal drawing, must have its own certificate of mailing or transmission.

Certificate of Mailing or Transmission

I hereby certify that this Fee(s) Transmittal is being deposited with the United States Postal Service with sufficient postage for first class mail in an envelope addressed to the Mail Stop ISSUE FEE address above, or being transmitted to the USPTO via EFS-Web or by facsimile to (571) 273-2885, on the date below.

(Typed or printed name)
(Signature)
(Date)

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

TITLE OF INVENTION: VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
nonprovisional	SMALL	$600	$0.00	$0.00	$600	03/18/2021

EXAMINER	ART UNIT	CLASS-SUBCLASS
LU, FRANK WEI MIN	1634	435-006110

1. Change of correspondence address or indication of "Fee Address" (37 CFR 1.363).

☐ Change of correspondence address (or Change of Correspondence Address form PTO/SB/122) attached.

☐ "Fee Address" indication (or "Fee Address" Indication form PTO/SB/47; Rev 03-09 or more recent) attached. **Use of a Customer Number is required.**

2. For printing on the patent front page, list
(1) The names of up to 3 registered patent attorneys or agents OR, alternatively,
(2) The name of a single firm (having as a member a registered attorney or agent) and the names of up to 2 registered patent attorneys or agents. If no name is listed, no name will be printed.

1 Gary J. Gershik
2
3

3. ASSIGNEE NAME AND RESIDENCE DATA TO BE PRINTED ON THE PATENT (print or type)

PLEASE NOTE: Unless an assignee is identified below, no assignee data will appear on the patent. If an assignee is identified below, the document must have been previously recorded, or filed for recordation, as set forth in 37 CFR 3.11 and 37 CFR 3.81(a). Completion of this form is NOT a substitute for filing an assignment.

(A) NAME OF ASSIGNEE: COLD SPRING HARBOR LABORATORY

(B) RESIDENCE: (CITY and STATE OR COUNTRY): COLD SPRING HARBOR, NEW YORK

Please check the appropriate assignee category or categories (will not be printed on the patent) : ☐ Individual ☒ Corporation or other private group entity ☐ Government

4a. Fees submitted: ☒ Issue Fee ☒ Publication Fee (if required) ☒ Advance Order - # of Copies Three (3)

4b. Method of Payment: *(Please first reapply any previously paid fee shown above)*

☐ Electronic Payment via EFS-Web ☐ Enclosed check ☐ Non-electronic payment by credit card (Attach form PTO-2038)

☒ The Director is hereby authorized to charge the required fee(s), any deficiency, or credit any overpayment to Deposit Account No. 03-3125

5. **Change in Entity Status** (from status indicated above)

☐ Applicant certifying micro entity status. See 37 CFR 1.29

☐ Applicant asserting small entity status. See 37 CFR 1.27

☐ Applicant changing to regular undiscounted fee status.

NOTE: Absent a valid certification of Micro Entity Status (see forms PTO/SB/15A and 15B), issue fee payment in the micro entity amount will not be accepted at the risk of application abandonment.
NOTE: If the application was previously under micro entity status, checking this box will be taken to be a notification of loss of entitlement to micro entity status.
NOTE: Checking this box will be taken to be a notification of loss of entitlement to small or micro entity status, as applicable.

NOTE: This form must be signed in accordance with 37 CFR 1.31 and 1.33. See 37 CFR 1.4 for signature requirements and certifications.

Authorized Signature /Gary J. Gershik/ Date January 21, 2021

Typed or printed name Gary J. Gershik Registration No. 39,992

PTOL-85 Part B (08-18) Approved for use through 01/31/2020 OMB 0651-0033 U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE
GUARDANT HEALTH EX 1009 692

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Darren Haber
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
PUBL. FEE- EARLY, VOLUNTARY, OR NORMAL	1504	1	0	0
UTILITY APPL ISSUE FEE	2501	1	600	600
Extension-of-Time:				
Miscellaneous:				
PRINTED COPY OF PATENT - NO COLOR	8001	3	3	9
		Total in USD ($)		**609**

Electronic Acknowledgement Receipt

EFS ID:	41710504
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Darren Haber
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	21-JAN-2021
Filing Date:	07-MAR-2016
Time Stamp:	17:24:48
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$609
RAM confirmation Number	E20211KH25020720
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.695

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Issue Fee Payment (PTO-85B)	210121_81503-AA_Issue_Fee-JGO.pdf	100841 2974f59da056063ba78dc88397fe7ea276965d46	no	1
Warnings:					
Information:					
2	Fee Worksheet (SB06)	fee-info.pdf	33869 20707ec00734c014445c9e32afe01c9d033970e9	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	134710		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

United States Patent and Trademark Office

Office of the Chief Financial Officer

Document Code:WFEE

User :C46539

Sale Accounting Date:01/25/2021

Sale Item Reference Number	Effective Date
15063278	01/21/2021

Document Number	Fee Code	Fee Code Description	Amount Paid	Payment Method
I20211O947053741	2830	PROCESSING FEE, EXCEPT PROV. APPLS.	$70.00	Deposit Account

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING or 371(c) DATE	GRP ART UNIT	FIL FEE REC'D	ATTY.DOCKET.NO	TOT CLAIMS	IND CLAIMS
15/063,278	03/07/2016	1634	730	81503-AA/GJG/AWG/AC	20	2

CONFIRMATION NO. 4038

UPDATED FILING RECEIPT

23432
COOPER & DUNHAM LLP
90 PARK AVENUE, 21 ST. FLOOR
NEW YORK, NY 10016

OC000000122873951

Date Mailed: 02/11/2021

Receipt is acknowledged of this non-provisional utility patent application. The application will be taken up for examination in due course. Applicant will be notified as to the results of the examination. Any correspondence concerning the application must include the following identification information: the U.S. APPLICATION NUMBER, FILING DATE, NAME OF FIRST INVENTOR, and TITLE OF INVENTION. Fees transmitted by check or draft are subject to collection.

Please verify the accuracy of the data presented on this receipt. If an error is noted on this Filing Receipt, please submit a written request for a corrected Filing Receipt, including a properly marked-up ADS showing the changes with strike-through for deletions and underlining for additions. If you received a "Notice to File Missing Parts" or other Notice requiring a response for this application, please submit any request for correction to this Filing Receipt with your reply to the Notice. When the USPTO processes the reply to the Notice, the USPTO will generate another Filing Receipt incorporating the requested corrections provided that the request is grantable.

Inventor(s)

James Hicks, Palm Desert, CA;
Nicholas Navin, Bellaire, TX;
Jennifer Clarke, Exeter, NH;
Zihua Wang, East Northport, NY;
Michael Wigler, Cold Spring Harbor, NY;

Applicant(s)

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY;

Assignment For Published Patent Application

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY

Power of Attorney: The patent practitioners associated with Customer Number 23432

Domestic Priority data as claimed by applicant

This application is a CON of 13/278,333 10/21/2011 PAT 9404156
which claims benefit of 61/510,579 07/22/2011
and claims benefit of 61/406,067 10/22/2010

Foreign Applications for which priority is claimed (You may be eligible to benefit from the **Patent Prosecution Highway** program at the USPTO. Please see http://www.uspto.gov for more information.) - None.
Foreign application information must be provided in an Application Data Sheet in order to constitute a claim to foreign priority. See 37 CFR 1.55 and 1.76.

GUARDANT HEALTH EX.1009.698

Permission to Access Application via Priority Document Exchange: Yes

Permission to Access Search Results: Yes

Applicant may provide or rescind an authorization for access using Form PTO/SB/39 or Form PTO/SB/69 as appropriate.

Projected Publication Date: Not Applicable
Non-Publication Request: No
Early Publication Request: No
** **SMALL ENTITY** **
Title

VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

Preliminary Class

435

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications: No

PROTECTING YOUR INVENTION OUTSIDE THE UNITED STATES

Since the rights granted by a U.S. patent extend only throughout the territory of the United States and have no effect in a foreign country, an inventor who wishes patent protection in another country must apply for a patent in a specific country or in regional patent offices. Applicants may wish to consider the filing of an international application under the Patent Cooperation Treaty (PCT). An international (PCT) application generally has the same effect as a regular national patent application in each PCT-member country. The PCT process **simplifies** the filing of patent applications on the same invention in member countries, but **does not result** in a grant of "an international patent" and does not eliminate the need of applicants to file additional documents and fees in countries where patent protection is desired.

Almost every country has its own patent law, and a person desiring a patent in a particular country must make an application for patent in that country in accordance with its particular laws. Since the laws of many countries differ in various respects from the patent law of the United States, applicants are advised to seek guidance from specific foreign countries to ensure that patent rights are not lost prematurely.

Applicants also are advised that in the case of inventions made in the United States, the Director of the USPTO must issue a license before applicants can apply for a patent in a foreign country. The filing of a U.S. patent application serves as a request for a foreign filing license. The application's filing receipt contains further information and guidance as to the status of applicant's license for foreign filing.

Applicants may wish to consult the USPTO booklet, "General Information Concerning Patents" (specifically, the section entitled "Treaties and Foreign Patents") for more information on timeframes and deadlines for filing foreign patent applications. The guide is available either by contacting the USPTO Contact Center at 800-786-9199, or it can be viewed on the USPTO website at http://www.uspto.gov/web/offices/pac/doc/general/index.html.

For information on preventing theft of your intellectual property (patents, trademarks and copyrights), you may wish to consult the U.S. Government website, http://www.stopfakes.gov. Part of a Department of Commerce initiative,

GUARDANT HEALTH EX.1009.699

this website includes self-help "toolkits" giving innovators guidance on how to protect intellectual property in specific countries such as China, Korea and Mexico. For questions regarding patent enforcement issues, applicants may call the U.S. Government hotline at 1-866-999-HALT (1-866-999-4258).

LICENSE FOR FOREIGN FILING UNDER

Title 35, United States Code, Section 184

Title 37, Code of Federal Regulations, 5.11 & 5.15

GRANTED

The applicant has been granted a license under 35 U.S.C. 184, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" followed by a date appears on this form. Such licenses are issued in all applications where the conditions for issuance of a license have been met, regardless of whether or not a license may be required as set forth in 37 CFR 5.15. The scope and limitations of this license are set forth in 37 CFR 5.15(a) unless an earlier license has been issued under 37 CFR 5.15(b). The license is subject to revocation upon written notification. The date indicated is the effective date of the license, unless an earlier license of similar scope has been granted under 37 CFR 5.13 or 5.14.

This license is to be retained by the licensee and may be used at any time on or after the effective date thereof unless it is revoked. This license is automatically transferred to any related applications(s) filed under 37 CFR 1.53(d). This license is not retroactive.

The grant of a license does not in any way lessen the responsibility of a licensee for the security of the subject matter as imposed by any Government contract or the provisions of existing laws relating to espionage and the national security or the export of technical data. Licensees should apprise themselves of current regulations especially with respect to certain countries, of other agencies, particularly the Office of Defense Trade Controls, Department of State (with respect to Arms, Munitions and Implements of War (22 CFR 121-128)); the Bureau of Industry and Security, Department of Commerce (15 CFR parts 730-774); the Office of Foreign AssetsControl, Department of Treasury (31 CFR Parts 500+) and the Department of Energy.

NOT GRANTED

No license under 35 U.S.C. 184 has been granted at this time, if the phrase "IF REQUIRED, FOREIGN FILING LICENSE GRANTED" DOES NOT appear on this form. Applicant may still petition for a license under 37 CFR 5.12, if a license is desired before the expiration of 6 months from the filing date of the application. If 6 months has lapsed from the filing date of this application and the licensee has not received any indication of a secrecy order under 35 U.S.C. 181, the licensee may foreign file the application pursuant to 37 CFR 5.15(b).

SelectUSA

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The U.S. offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to promote and facilitate business investment. SelectUSA provides information assistance to the international investor community; serves as an ombudsman for existing and potential investors; advocates on behalf of U.S. cities, states, and regions competing for global investment; and counsels U.S. economic development organizations on investment attraction best practices. To learn more about why the United States is the best country in the world to develop

technology, manufacture products, deliver services, and grow your business, visit http://www.SelectUSA.gov or call +1-202-482-6800.

GUARDANT HEALTH EX.1009.701

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NUMBER	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTY. DOCKET NO./TITLE
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC

CONFIRMATION NO. 4038

23432
COOPER & DUNHAM LLP
90 PARK AVENUE, 21 ST. FLOOR
NEW YORK, NY 10016

37 CFR 1.48(f)
ACKNOWLEDGEMENT LETTER

OC000000122873953

Date Mailed: 02/11/2021

NOTICE OF ACCEPTANCE OF REQUEST UNDER 37 CFR 1.48(f)

This is in response to the applicant's request under 37 CFR 1.48(f) submitted on 01/21/2021.

The request under 37 CFR 1.48(f) to correct the inventorship, to correct or update the name of an inventor, or to correct the order of names of joint inventors is accepted.

Questions about the contents of this notice and the requirements it sets forth should be directed to the Office of Data Management, Application Assistance Unit, at **(571) 272-4000** or **(571) 272-4200** or **1-888-786-0101**.

/ttran/

GUARDANT HEALTH EX.1009.702

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	ISSUE DATE	PATENT NO.	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/16/2021	10947589	81503-AA/GJG/AWG/AC	4038

23432 7590 02/24/2021

COOPER & DUNHAM LLP
90 PARK AVENUE, 21 ST. FLOOR
NEW YORK, NY 10016

ISSUE NOTIFICATION

The projected patent number and issue date are specified above.

Determination of Patent Term Adjustment under 35 U.S.C. 154 (b)
(application filed on or after May 29, 2000)

The Patent Term Adjustment is 45 day(s). Any patent to issue from the above-identified application will include an indication of the adjustment on the front page.

If a Continued Prosecution Application (CPA) was filed in the above-identified application, the filing date that determines Patent Term Adjustment is the filing date of the most recent CPA.

Applicant will be able to obtain more detailed information by accessing the Patent Application Information Retrieval (PAIR) WEB site (http://pair.uspto.gov).

Any questions regarding the Patent Term Extension or Adjustment determination should be directed to the Office of Patent Legal Administration at (571)-272-7702. Questions relating to issue and publication fee payments should be directed to the Application Assistance Unit (AAU) of the Office of Data Management (ODM) at (571)-272-4200.

APPLICANT(s) (Please see PAIR WEB site http://pair.uspto.gov for additional applicants):

James Hicks, Palm Desert, CA;
Cold Spring Harbor Laboratory, Cold Spring Harbor, NY;
Nicholas Navin, Bellaire, TX;
Jennifer Clarke, Exeter, NH;
Zihua Wang, East Northport, NY;
Michael Wigler, Cold Spring Harbor, NY;

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The USA offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to encourage and facilitate business investment. To learn more about why the USA is the best country in the world to develop technology, manufacture products, and grow your business, visit SelectUSA.gov.

GUARDANT HEALTH EX.1009.703

DocCode - PATENT.GRANT

SCORE Placeholder Sheet for IFW Content

Application Number: 15063278 Document Date: 03/16/2021

The presence of this form in the IFW record indicates that the following document type was received in electronic format on the date identified above. This content is stored in the SCORE database.

Since this was an electronic submission, there is no physical artifact folder, no artifact folder is recorded in PALM, and no paper documents or physical media exist. The TIFF images in the IFW record were created from the original documents that are stored in SCORE.

- Patent Grant with Color/Grayscale Drawings

At the time of document entry (noted above):

- USPTO employees may access SCORE content via DAV or via the SCORE web page.
- External customers may access SCORE content via PAIR using the Supplemental Content tab.

Form Revision Date: March 1, 2019

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	: Cold Spring Harbor Laboratory
U.S. Patent No.	: 10,947,589
Issue Date	: March 16, 2021
U.S. Serial No.	: 15/063,278
Filing Date	: March 7, 2016
Confirmation No.	: 4038
For	: VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION

90 Park Avenue
21st Floor
New York, NY 10016
May 13, 2021

BY EFS
Commissioner for Patents
Alexandria, VA 22313-1450

PETITION REQUESTING RECONSIDERATION OF PATENT TERM ADJUSTMENT

Pursuant to 37 C.F.R. §1.705, Applicant/Patentee hereby requests reconsideration and recalculation of the Patent Term Adjustment ("PTA") printed on the face of above-identified U.S. Patent No. 10,947,589 ("the '589 Patent"). According to 37 C.F.R. §1.705(b) a request for reconsideration of patent term adjustment must be filed no later than two months from the date the patent was granted. The '589 Patent was granted on March 16, 2021. Accordingly, the deadline to file a request is May 17, 2021 (because May 16, 2021 is a Sunday) and this petition is being timely filed.

In accordance with 37 C.F.R. §1.705 patentee provides herewith the fee set forth in 37 C.F.R. 1.18(e) and a statement setting for the facts involved specifying:

(i) The correct patent term adjustment and the basis or bases under § 1.702 for the adjustment;

(ii) The relevant dates as specified in §§ 1.703(a) through (e) for which an adjustment is sought and the adjustment as specified in § 1.703(f) to which the patent is entitled;

(iii) Whether the patent is subject to a terminal disclaimer and any expiration date specified in the terminal disclaimer; and

(iv) (A) Any circumstances during the prosecution of the application resulting in the patent that constitute a failure to engage in reasonable efforts to conclude processing or examination of such application as set forth in § 1.704; or

(B) That there were no circumstances constituting a failure to engage in reasonable efforts to conclude processing or examination of such application as set forth in § 1.704.

(i) The correct patent term adjustment and the basis or bases under § 1.702 for the adjustment

The correct patent term adjustment is 79 days. This adjustment is based on the bases under 37 C.F.R. § 1.702 presented in this section, the cumulative total of Office delay as discussed in section (ii) below, and the cumulative total of applicant delay as discussed in section (iv) below.

37 C.F.R. § 1.702(a)(1)

Under 37 C.F.R. § 1.702(a)(1) the Office did not mail at least one of a notification under 35 U.S.C. 132 or a notice of allowance under 35 U.S.C. 151 not later than fourteen months after the date on which the application was filed under 35 U.S.C. 111(a). The application was filed under 35 U.S.C. 111(a) on March 7, 2016 and a first office action as mailed on January 18, 2018, a delay of 256 days.

37 C.F.R. § 1.702(a)(2)

Under 37 C.F.R. § 1.702(a)(2) the Office did not respond to a reply under 35 U.S.C. 132 or to an appeal taken under 35 U.S.C. 134 not later than four months after the date on which the reply was filed or the appeal was taken for a cumulative total of 77 days of Office delay as discussed in section (ii) below.

37 C.F.R. § 1.702(b)

Under 37 C.F.R. § 1.702(b) the term of an original patent shall be adjusted if the issuance of the patent was delayed due to the failure of the Office to issue a patent within three years after the date on which the application was filed under 35 U.S.C. 111(a) or the national stage commenced under 35 U.S.C. 371(b) or (f) in an international application. The application was filed under 35 U.S.C. 111(a) on March 7, 2016 and issued March 16, 2021, a delay of 739 days.

With respect to the ground for adjustment set forth in 37 C.F.R. § 1.702(b)(1), the period of adjustment set forth in 37 C.F.R. § 1.703 shall be reduced by the number of days consumed by continued examination of the application under 35 U.S.C. 132(b). A Request for Continued Examination was filed on May 30, 2019 and a Notice of Allowance was mailed December 18, 2020. The number of days to exclude for continued examination is 568 days.

With respect to the grounds for adjustment set forth in 37 C.F.R. § 1.702(b)(5), the period of adjustment set forth in 37 C.F.R. § 1.703 shall be reduced by the number of days of any delay in the processing of the application by the Office that was requested by the applicant. The cumulative total 425 days of applicant delay is discussed in section (iv) below.

(ii) The relevant dates as specified in §§ 1.703(a) through (e) for which an adjustment is sought and the adjustment as specified in § 1.703(f) to which the patent is entitled

The relevant dates as specified in §§ 1.703(a) through (e) for which an adjustment is sought are provided below:

37 C.F.R. § 1.703(a)(1)

Under 37 C.F.R. § 1.703(a)(1), the period of adjustment is the number of days in the period beginning on the day after the date that is fourteen months after the date on which the application was filed under 35 U.S.C. 111(a) or the date the national stage commenced under 35 U.S.C. 371(b) or (f) in an international application and ending on the date of mailing of either an action under 35 U.S.C. 132, or a notice of allowance under 35 U.S.C. 151, whichever occurs first.

The application was filed under 35 U.S.C. 111(a) on March 7, 2016. The date that is fourteen months after the date on which the application was filed is May 7, 2017, and the day after this date is May 8, 2017. The mailing of an action under 35 U.S.C. 132 occurred on January 18, 2018. This period of delay entitles the application to 256 days of patent term adjustment.

37 C.F.R. § 1.703(a)(2)

Under 37 C.F.R. § 1.703(a)(2), the period of adjustment is the number of days in the period beginning on the day after the date that is four months after the date a reply under § 1.111 was filed and ending on the date of mailing of either an action under 35 U.S.C. 132, or a notice of allowance under 35 U.S.C. 151, whichever occurs first.

A reply under § 1.111 was filed on May 30, 2019. The date that is four months after the date on which the reply was filed is September 30, 2019, and the day after this date is October 1, 2019. The mailing of an action under 35 U.S.C. 132 occurred on October 3, 2019. This period of delay entitles the application to 3 days of patent term adjustment.

A reply under § 1.111 was filed on April 3, 2020. The date that is four months after the date on which the reply was filed is August 3, 2020, and the day after this date is August 4, 2020. The mailing of an action under 35 U.S.C. 132 occurred on October 16, 2020. This period of delay entitles the application to 74 days of patent term adjustment.

37 C.F.R. § 1.703(b)

Under 37 C.F.R. § 1.703(b), the period of adjustment is the number of days in the period beginning on the day after the date that is three years after the date on which the application was filed under 35 U.S.C. 111(a) or the national stage commenced under 35 U.S.C. 371(b) or (f) in an international application and ending on the date a patent was issued. The application was filed under 35 U.S.C. 111(a) on March 7, 2016. The date that is three years after the date on which the application was filed is March 7, 2019, and the day after this date is March 8, 2019. The date a patent was issued is March 16, 2021. This period of delay entitles the application to 739 days of patent term adjustment.

Under 37 C.F.R. § 1.703(b)(1), the period of adjustment does not include the sum of the number of days in the period beginning on the date on which any request for continued examination of the application under 35 U.S.C. 132(b) was filed and ending on the date of mailing of the notice of allowance under 35 U.S.C. 151. A Request for Continued Examination was filed on May 30, 2019, and a Notice of Allowance was mailed December 18, 2020. This period of delay reduces the period of adjustment by 568 days.

37 C.F.R. §§ 1.703(c)-(e)

No adjustment is sought under these subsections.

37 C.F.R. § 1.703(f)

Under 37 C.F.R. § 1.703(f) "[t]he term of a patent entitled to adjustment under § 1.702 and this section shall be adjusted for the sum of the periods calculated under paragraphs (a) through (e) of this section, to the extent that such periods are not overlapping, less the sum of the periods calculated under § 1.704."

None of adjustment under § 1.702 is overlapping. Accordingly, the period of adjustment as specified in § 1.703(f) to which the patent is entitled is 333 days under § 1.703(a) and 171 days under § 1.703(b), less the sum of periods calculated under § 1.704.

As noted in section (iv) below, the sum of the periods calculated under § 1.704 is 425. Accordingly, the period of adjustment as specified in § 1.703(f) to which the patent is entitled is 79 days.

(iii) Whether the patent is subject to a terminal disclaimer and any expiration date specified in the terminal disclaimer

The above-identified patent is subject to a terminal disclaimer due to the issuance of U.S. Patent No. 9,404,156, issued August 2, 2016, which, with its 731 days of PTA will expire on October 21, 2033.

(iv) Any circumstances during the prosecution of the application resulting in the patent that constitute a failure to engage in reasonable efforts to conclude processing or examination of such application as set forth in § 1.704

The automated PTA calculation in PAIR assigns 459 days of applicant delay in placing the application in condition for examination, which appears to be based on 37 C.F.R. §§ 1.704(b), 1.704(c)(8), and 1.704(c)(10). Applicant/patentee calculates 425 days of applicant delay based on 37 C.F.R. §§ 1.704(b), 1.704(c)(8), and 1.704(c)(10).

37 C.F.R. § 1.704(b)

Under 37 C.F.R. §1.704(b), the period of adjustment set forth in §1.703 shall be reduced by the number of days beginning on the day after the date that is three months after the date of mailing of the Office communication and ending on the date the reply was filed.

An Office communication was mailed on April 17, 2018. The date that is three months after the date on which the reply was filed is July 17, 2018, and the day after this date is July 18, 2018. A reply was filed on October 17, 2018. This period of delay reduces the period of adjustment by 92 days.

An Office communication was mailed on November 30, 2018. The date that is three months after the date on which the reply was filed is February 28, 2019, and the day after this date is March 1, 2019. A reply was filed on May 30, 2019. This period of delay reduces the period of adjustment by 91 days.

An Office communication was mailed on October 3, 2019. The date that is three months after the date on which the reply was filed is January 3, 2020, and the day after this date is January 4, 2020. A reply was filed on April 3, 2020. This period of delay reduces the period of adjustment by 91 days.

37 C.F.R. § 1.704(c)(8)

Under 37 C.F.R. §1.704(c)(8), the period of adjustment set forth in §1.703 shall be reduced by the number of days beginning on the day after the date the initial reply was filed and ending on the date that the submission of a supplemental reply or other paper, other than a supplemental reply or other paper expressly requested by the examiner, was filed.

A reply was filed April 3, 2020. The date that is after this date is April 4, 2020. A supplemental reply was filed on August 11, 2020. This period of delay reduces the period of adjustment by 130 days.

37 C.F.R. § 1.704(c)(10)

Under 37 C.F.R. §1.704(c)(10), the period of adjustment set forth in §1.703 shall be reduced by the lesser of (i) the number of days beginning on the date the amendment under § 1.312 or other paper, other than a request for continued examination in compliance with § 1.114, after a notice of allowance has been given or mailed was filed and ending on the mailing date of the Office action or notice in response to the amendment under § 1.312 or such other paper or (ii) four months.

An amendment under § 1.312 was filed January 21, 2021. A notice in response to the amendment under § 1.312, specifically a Notice of Acceptance of Request under 37 CFR 1.48(f), was mailed February 11, 2021. Accordingly, this period of delay reduces the period of adjustment by 21 days.

Total Days under 37 C.F.R. § 1.704

Applicant/patentee calculates the cumulative applicant delay under 37 C.F.R. § 1.704 is 425 days.

Conclusion

As set forth above, the '589 patent is entitled to 504 days of PTA under 37 C.F.R. §§1.702 and 1.703. This PTA is reduced by 425 days of applicant delay pursuant to 37 C.F.R. §1.704. Therefore, the correct PTA for the '589 patent is 79 days.

Pursuant to 37 C.F.R. § 1.705, appropriate correction of the PTA shown on the face of the patent is respectfully requested.

No fee, other than the fee of $210.00 under 37 C.F.R. § 1.18(e), is deemed necessary in connection with the filing of this Petition. However, if any additional fee is required, authorization is hereby given to charge the amount of such fee to Deposit Account No. 03-3125.

Respectfully submitted,

/Gary J. Gershik/

Gary J. Gershik, Esq.
Registration No. 39,992
Attorney for Applicant
Cooper & Dunham LLP (Customer #23432)
90 Park Avenue
21st Floor
New York, NY 10016
(212) 278-0400

Certificate of Transmission

I hereby certify that this correspondence is being transmitted via the Electronic Filing System (EFS) to the U.S. Patent and Trademark Office on May 13, 2021.

/Lourania M. Oliver/

Lourania M. Oliver

Electronic Patent Application Fee Transmittal

Application Number:	15063278
Filing Date:	07-Mar-2016
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Filer:	Gary J. Gershik/Brittany Internoscia
Attorney Docket Number:	81503-AA/GJG/AWG/AC

Filed as Small Entity

Filing Fees for Utility under 35 USC 111(a)

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Basic Filing:				
Pages:				
Claims:				
Miscellaneous-Filing:				
Petition:				
APPLICATION FOR PATENT TERM ADJUSTMENT	2455	1	210	210
Patent-Appeals-and-Interference:				
Post-Allowance-and-Post-Issuance:				

Description	Fee Code	Quantity	Amount	Sub-Total in USD($)
Extension-of-Time:				
Miscellaneous:				
			Total in USD ($)	**210**

Electronic Acknowledgement Receipt

EFS ID:	42717551
Application Number:	15063278
International Application Number:	
Confirmation Number:	4038
Title of Invention:	VARIETAL COUNTING OF NUCLEIC ACIDS FOR OBTAINING GENOMIC COPY NUMBER INFORMATION
First Named Inventor/Applicant Name:	James Hicks
Customer Number:	23432
Filer:	Gary J. Gershik/Brittany Internoscia
Filer Authorized By:	Gary J. Gershik
Attorney Docket Number:	81503-AA/GJG/AWG/AC
Receipt Date:	13-MAY-2021
Filing Date:	07-MAR-2016
Time Stamp:	15:57:58
Application Type:	Utility under 35 USC 111(a)

Payment information:

Submitted with Payment	yes
Payment Type	DA
Payment was successfully received in RAM	$210
RAM confirmation Number	E20215CF58132938
Deposit Account	
Authorized User	

The Director of the USPTO is hereby authorized to charge indicated fees and credit any overpayment as follows:

GUARDANT HEALTH EX.1009.715

File Listing:

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Patent Term Adjustment Petition	210513_81503-AA_PTARequest_LMO.pdf	112433 65686e57f7044b6610f9ea290ab4258ad7027a5f	no	8
Warnings:					
Information:					
2	Fee Worksheet (SB06)	fee-info.pdf	30464 953146a5fe5adf72d4ac217a8470398158f467ba	no	2
Warnings:					
Information:					
		Total Files Size (in bytes):	142897		

This Acknowledgement Receipt evidences receipt on the noted date by the USPTO of the indicated documents, characterized by the applicant, and including page counts, where applicable. It serves as evidence of receipt similar to a Post Card, as described in MPEP 503.

New Applications Under 35 U.S.C. 111

If a new application is being filed and the application includes the necessary components for a filing date (see 37 CFR 1.53(b)-(d) and MPEP 506), a Filing Receipt (37 CFR 1.54) will be issued in due course and the date shown on this Acknowledgement Receipt will establish the filing date of the application.

National Stage of an International Application under 35 U.S.C. 371

If a timely submission to enter the national stage of an international application is compliant with the conditions of 35 U.S.C. 371 and other applicable requirements a Form PCT/DO/EO/903 indicating acceptance of the application as a national stage submission under 35 U.S.C. 371 will be issued in addition to the Filing Receipt, in due course.

New International Application Filed with the USPTO as a Receiving Office

If a new international application is being filed and the international application includes the necessary components for an international filing date (see PCT Article 11 and MPEP 1810), a Notification of the International Application Number and of the International Filing Date (Form PCT/RO/105) will be issued in due course, subject to prescriptions concerning national security, and the date shown on this Acknowledgement Receipt will establish the international filing date of the application.

UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	4038

23432 7590 01/13/2022

Gary J. Gershik; John P. White;
Christopher C. Dunham; Paul Teng
90 PARK AVENUE, 21st Fl
NEW YORK, NY 10016

EXAMINER	
LU, FRANK WEI MIN	

ART UNIT	PAPER NUMBER
1634	

NOTIFICATION DATE	DELIVERY MODE
01/13/2022	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

PTODOCKET@ARELAW.COM
klees@ARELAW.com
patcomm-in-grp@cooperdunham.com

GUARDANT HEALTH EX.1009.717

UNITED STATES PATENT AND TRADEMARK OFFICE

Commissioner for Patents
United States Patent and Trademark Office
P.O. Box 1450
Alexandria, VA 22313-1450
www.uspto.gov

In re Patent No. 10,947,589		
Issue Date: 16 Mar 2021	:	
Application No. 15/063,278	:	
Filing or 371(c) Date: 7 Mar 2016	:	REDETERMINATION OF
For: VARIETAL COUNTING OF	:	PATENT TERM ADJUSTMENT
NUCLEIC ACIDS FOR OBTAINING	:	
GENOMIC COPY NUMBER	:	
INFORMATION	:	

This is a decision on patentee's "REQUEST FOR RECONSIDERATION OF PATENT TERM ADJUSTMENT" filed May 13, 2021, requesting the Office adjust the patent term adjustment (PTA) from 45 days to 79 days.

The Office has re-determined the PTA to be **66 days**.

This redetermination of patent term adjustment is NOT the Director's decision on the patentee's request for reconsideration for purposes of seeking judicial review under 35 U.S.C. § 154(b)(4).

Relevant Procedural History

On March 16, 2021, this patent issued with a PTA of 45 days. On May 13, 2021, patentee filed the present request for redetermination of patent term adjustment seeking 79 days of patent term adjustment.

Decision

The PTA is the sum of the days of A Delay, B Delay, and C Delay reduced by the number of days of Overlap and Applicant Delay. In other words, the following formula is be used to calculate the PTA:

PTA = A Delay + B Delay + C Delay – Overlap – Applicant Delay

The patent set forth a PTA of 45 days based on the following:

(1) A period of delay under 35 U.S.C. 154(b)(1)(A) ("A Delay") of 333 days;
(2) A period of delay under 35 U.S.C. 154(b)(1)(B) ("B Delay") of 171 days;
(3) The period of delay under 35 U.S.C. 154(b)(1)(C) ("C Delay") of 0 days;
(4) The number of days of overlapping delay ("Overlap") between the periods of A Delay, B Delay, and C Delay of 0 days; and
(5) The period of delay under 35 U.S.C. 154(b)(2)(C) ("Applicant Delay") as 459 days.

333 days of A Delay + 171 days of B Delay + 0 days of C Delay - 0 days of Overlap – 459 days = 45 days.

The Office notes that final rule changes set forth in *Patent Term Adjustment Reductions in View of the Federal Circuit Decision in Supernus Pharm., Inc. v. Iancu*, 85 FR 36335 (June 16, 2020) apply to original utility an plant patents issuing from applications filed on or after May 29, 2000, in which a Notice of Allowance was mailed on or after July 16, 2020. The Office notes that utility patent No. 10,947,589 issued from an application filed on March 7, 2016, after May 29, 2000, and where a Notice of Allowance was mailed on December 18, 2020, after July 16, 2020. Therefore, the Office will apply the rule changes set forth in *Supernus* when issuing its redetermination on PTA.

The Office has determined the correct amount of PTA is **66 days** (333 days of A Delay + 171 days of B Delay + 0 days of C Delay - 0 days of Overlap – 438 days of Applicant Delay).

"A" Delay

Patentee and the Office agree the amount of "A" delay is 333 days.

"B" Delay

Patentee and the Office agree about the amount of "B" delay is **171 days**.

"C" Delay

Patentee and the Office agree that the amount of "C" delay under 37 CFR 1.703(e) is **0 days**.

Overlap

Patentee and the Office agree the amount of overlapping days between the "A" delay and "B" delay periods is **0 days**.

Applicant Delay

Patentee and the Office disagree about the amount of "B" delay.

Patentee disputes the Office's prior determination of 459 days of applicant delay. Patentee avers the correct amount of applicant delay is 425 days. In particular, patentee solely disagrees with the Office's calculation of the period of reduction for the filing an amendment under § 1.312 on January 12, 2021, after the mailing of the Notice of Allowance. Patentee maintains the applicant delay under 37 CFR 1.704(c)(10) is 21 days, the number of days beginning on January 12, 2021 (date the amendment under § 1.312 was filed), and ending on February 11, 2021 (date the Notice of Acceptance under 37 CFR 1.48(f) was mailed).

As stated above, the Office will apply the rule changes set forth in *Supernus* when issuing its redetermination on PTA.

37 CFR 1.704(c)(10) states,

> Submission of an amendment under § 1.312 or other paper, other than an amendment under § 1.312 or other paper expressly requested by the Office or a request for continued examination in compliance with § 1.114, after a notice of allowance has been given or mailed, in which case the period of adjustment set forth in § 1.703 shall be reduced by the number of days, if any, beginning on the day after the date of mailing of the notice of allowance under 35 U.S.C. 151 and ending on the date the amendment under § 1.312 or other paper was filed;

The Office has recalculated the number of days of applicant delay as part of the Office's redetermination of the PTA and found applicant delay is **438** days.

Applicant delay includes the following period:

(1) 92 days of applicant delay under 37 CFR 1.704(b) for the number of days beginning on July 18, 2018 (day after the date that is three months after the date of mailing or transmission of the Office communication notifying the applicant of the rejection, objection, argument, or other request) and ending on October 17, 2018 (date the reply was filed)
(2) 91 days of applicant delay under 37 CFR 1.704(b) for the number of days beginning on March 1, 2019 (day after the date that is three months after the date of mailing or transmission of the Office communication notifying the applicant of the rejection, objection, argument, or other request) and ending on May 30, 2019 (date the reply was filed)
(3) 91 days of applicant delay under 37 CFR 1.704(b) for the number of days beginning on January 4, 2020 (day after the date that is three months after the date of mailing or transmission of the Office communication notifying the applicant of the rejection, objection, argument, or other request) and ending on April 3, 2020 (date the reply was filed)

(4) 130 days of applicant delay under 37 CFR 1.704(c)(8) for the number of days beginning on April 4, 2020 (day after the date the initial reply was filed) and ending on August 11, 2020 (date the supplemental reply or other paper, other than a supplemental reply or other paper was filed)
(5) 34 days of applicant delay under 37 CFR 1.704(c)(10) for the number of days beginning on December 19, 2020 (day after the date of mailing date of the notice of allowance under 35 U.S.C. 151) and ending on January 21, 2021 (date the amendment under § 1.312 or other paper was filed).

Consistent with the rule change to 37 CFR 1.704(c)(10), the Office has determined the amount of applicant delay for the filing of the amendment under § 1.312 on January 21, 2021, after the mailing of the Notice of Allowance on December 18, 2020, is 34 days. Accordingly, the Office will remove the period of reduction of 55 days and enter a period of reduction of 34 days.

The Office finds applicant delay is **438** days.

Overall PTA Calculation

Formula:

"A" delay + "B" delay + "C" delay - Overlap - Applicant delay = X days of PTA

USPTO's Calculation:

333 + 171 + 0 – 0 – 438= 66 days

Patentee's Calculation:

333 + 171 + 0 – 0 – 425 = 79 days

Conclusion

Patentee is entitled to PTA of 66 days. Using the formula "A" delay + "B" delay + "C" delay - Overlap - Applicant delay = X, the amount of PTA is calculated as follows: 333 + 171 + 0 – 0 – 438 = 79 days.

If patentee continues to disagree with the Office's redetermination of patent term adjustment, patentee has two (2) months from the date of this redetermination to request reconsideration of the patent term adjustment without paying any additional petition fee. This two-month period is extendable under 37 CFR 1.136(a). However, if patentee responds more than two months after the mail date of the redetermination, patentee is required to pay the extension of time fee. After

the period to respond has expired, the Office will *sua sponte* issue a certificate of correction adjusting the PTA to **66** days.

Questions related to this matter may be directed to the undersigned at (571) 272-3211.

/CHRISTINA TARTERA DONNELL/
Attorney Advisor, OPET

Enclosures: Adjusted PTA calculation
Draft Certificate of Correction

Patent Term Adjustments

Patent Term Adjustment (PTA) for Application Number: 15/063,278

Filing or 371(c) Date:	03-07-2016	Overlapping Days Between {A and B} or {A and C}:	0
Issue Date of Patent:	03-16-2021	Non-Overlapping USPTO Delays:	504
A Delays:	333	PTO Manual Adjustments:	21
B Delays:	171	Applicant Delays:	459
C Delays:	0	Total PTA Adjustments:	66

Patent Term Adjustment History | Explanation Of Calculations

Number	Date	Contents Description	PTO(Days)	APPL(Days)	Start
165	01-13-2022	Adjustment of PTA Calculation by PTO		34	0
164	01-13-2022	Adjustment of PTA Calculation by PTO	55		0
158.5	03-16-2021	PTA 36 Months	171		0.5
158	03-16-2021	Patent Issue Date Used in PTA Calculation			0
157	02-12-2021	Export to Final Data Capture			0
156	02-11-2021	Dispatch to FDC			0
155	02-11-2021	Email Notification			0
154	02-11-2021	Email Notification			0
153	02-11-2021	Letter Accepting Correction of Inventorship Under Rule 1.48			0
152	02-11-2021	Filing Receipt - Updated			0
151	01-25-2021	Finished Initial Data Capture			0
150	01-21-2021	Supplemental Papers - Oath or Declaration			0
149	01-21-2021	Miscellaneous Incoming Letter		55	158
148	01-22-2021	Application Is Considered Ready for Issue			0
147	01-21-2021	Issue Fee Payment Verified			0
146	01-21-2021	Issue Fee Payment Received			0
145	12-23-2020	Sequence Forwarded to Pubs on Tape			0
144	12-18-2020	Export to Initial Data Capture			0
143	12-18-2020	Electronic Review			0
142	12-18-2020	Email Notification			0

141	12-18-2020	Mail Notice of Allowance	0
140	12-16-2020	Office Action Review	0
139	12-16-2020	Office Action Review	0
138	12-16-2020	Office Action Review	0
137	12-16-2020	Office Action Review	0
136	12-16-2020	Office Action Review	0
135	12-16-2020	Issue Revision Completed	0
134	12-15-2020	Office Action Review	0
133	12-15-2020	Office Action Review	0
132	12-15-2020	Office Action Review	0
131	12-15-2020	Office Action Review	0
130	12-15-2020	Office Action Review	0
129	12-15-2020	Allowed Case Returned to the Examiner for Clerical Processing	0
128	12-15-2020	Notice of Allowance Data Verification Completed	0
127	12-15-2020	Document Verification	0
126	12-14-2020	After Final Consideration Program Additional Consideration and/or updated search	0
125	12-14-2020	Examiner's Amendment Communication	0
124	12-14-2020	Interview Summary - Examiner Initiated - Telephonic	0
123	12-14-2020	Reasons for Allowance	0
122	12-14-2020	Allowability Notice	0
121	12-02-2020	Date Forwarded to Examiner	0
120	11-25-2020	Response after Final Action	0
119	12-01-2020	Email Notification	0
118	12-01-2020	Mail Examiner Interview Summary (PTOL - 413)	0
117	11-24-2020	Interview Summary - Applicant Initiated - Telephonic	0
116	11-24-2020	Interview Summary Record	0
115	11-20-	Email Notification	0

	2020				
114	11-20-2020	Mail Examiner Interview Summary (PTOL - 413)			0
113	11-17-2020	Interview Summary - Examiner Initiated - Telephonic			0
112	11-17-2020	Interview Summary Record			0
111	10-19-2020	Office Action Review			0
110	10-16-2020	Electronic Review			0
109	10-16-2020	Misc Special Soft Scanning- No Mailing			0
108	10-16-2020	Email Notification			0
107	10-16-2020	Mail Final Rejection (PTOL - 326)	74		88
106	10-11-2020	Final Rejection			0
105	10-11-2020	Information Disclosure Statement considered			0
104	08-13-2020	Date Forwarded to Examiner			0
103	08-11-2020	Supplemental Response		130	88
99	06-18-2020	Date Forwarded to Examiner			0
98	06-04-2020	Supplemental Response			0
97	06-15-2020	Email Notification			0
96	06-12-2020	Mail Examiner Initiated Interview Summary			0
95	06-08-2020	Interview Summary - Examiner Initiated - Telephonic			0
94	06-08-2020	Interview Summary - Examiner Initiated			0
93	05-18-2020	Mail-Petition Decision - Granted			0
92	10-17-2018	Petition Decision - Granted			0
91	03-07-2016	Petition Entered			0
90	04-03-2020	Miscellaneous Incoming Letter			0
89	04-07-2020	Date Forwarded to Examiner			0
88	04-03-2020	Response after Non-Final Action		91	84
87	04-03-2020	Request for Extension of Time - Granted			0
86	04-03-2020	Information Disclosure Statement (IDS) Filed			0

85	04-03-2020	Information Disclosure Statement (IDS) Filed			0
84	10-03-2019	Mail Non-Final Rejection	3		77
83	10-01-2019	Add FAOM 1.25 Count			0
82	10-01-2019	Subtract FAOM 1.00 Count			0
81	10-01-2019	Non-Final Rejection			0
80	09-30-2019	Information Disclosure Statement considered			0
79	06-03-2019	Date Forwarded to Examiner			0
78	05-30-2019	RCE- AF Processed			0
77	05-30-2019	Request for Continued Examination (RCE)		91	59
76	06-03-2019	Disposal for a RCE / CPA / R129			0
75	05-30-2019	Workflow - Request for RCE - Begin			0
74	04-25-2019	Mail Advisory Action (PTOL - 303)			0
73	04-25-2019	PARALEGAL OR ELECTRONIC TERMINAL DISCLAIMER APPROVED			0
72	04-17-2019	Terminal Disclaimer Filed			0
71	04-24-2019	After Final Consideration Program Additional Consideration and/or updated search			0
70	04-24-2019	Interview Summary - Examiner Initiated - Telephonic			0
69	04-24-2019	Advisory Action (PTOL-303)			0
68	04-17-2019	PILOT- Request for After Final Consideration Program			0
67	04-19-2019	Date Forwarded to Examiner			0
66	04-17-2019	Response after Final Action			0
65	04-17-2019	Request for Extension of Time - Granted			0
64	04-17-2019	Incoming Letter Pertaining to the Drawings			0
63	02-25-2019	Information Disclosure Statement (IDS) Filed			0
62	04-19-2019	Filing Receipt - Corrected			0
61	04-19-2019	Change in Power of Attorney (May Include Associate POA)			0
60	02-25-2019	Information Disclosure Statement (IDS) Filed			0

59	11-30-2018	Mail Final Rejection (PTOL - 326)			0
58	11-29-2018	Final Rejection			0
57	11-04-2018	Date Forwarded to Examiner			0
56	10-17-2018	Response after Non-Final Action		92	48
55	10-17-2018	Request for Extension of Time - Granted			0
54	11-02-2018	Mail-Petition Decision - Granted			0
53	11-02-2018	Petition Decision - Granted			0
52	10-17-2018	Petition Entered			0
51	10-17-2018	Mail TC Petition Decision			0
50	10-17-2018	Office Action Review			0
49	10-16-2018	TC Petition Decision			0
48	04-17-2018	Mail Non-Final Rejection			0
47	04-16-2018	Office Action Review			0
46	04-14-2018	Non-Final Rejection			0
45	04-13-2018	Information Disclosure Statement considered			0
44	04-13-2018	Information Disclosure Statement considered			0
43	04-13-2018	Information Disclosure Statement considered			0
42	03-24-2018	Date Forwarded to Examiner			0
41	03-16-2018	Response to Election / Restriction Filed			0
40	03-16-2018	Information Disclosure Statement (IDS) Filed			0
39	03-16-2018	Information Disclosure Statement (IDS) Filed			0
38	01-18-2018	Mail Restriction Requirement	256		0.5
37	01-14-2018	Office Action Review			0
36	01-09-2018	Restriction/Election Requirement			0
33	09-06-2017	Information Disclosure Statement (IDS) Filed			0
32	09-06-2017	Information Disclosure Statement (IDS) Filed			0
31	09-06-	Information Disclosure Statement (IDS) Filed			0

	2017		
30	06-19-2017	Preliminary Amendment	0
29	09-13-2016	Preliminary Amendment	0
27	09-01-2016	PG-Pub Issue Notification	0
26	07-13-2016	Mail Non-Compliant Preliminary Amendment	0
25	03-08-2016	Preliminary Amendment	0
24	07-12-2016	Non-Compliant Preliminary Amendment	0
23	03-07-2016	Preliminary Amendment	0
22	05-31-2016	Case Docketed to Examiner in GAU	0
21	05-24-2016	CRF Is Good Technically / Entered into Database	0
20	05-25-2016	Application Dispatched from OIPE	0
19	05-25-2016	FITF set to NO - revise initial setting	0
18	05-20-2016	Patent Term Adjustment - Ready for Examination	0
17	05-25-2016	Application Is Now Complete	0
16	05-25-2016	Filing Receipt - Updated	0
15	05-25-2016	Application Is Now Complete	0
14	05-20-2016	Additional Application Filing Fees	0
13	05-20-2016	A set of symbols and procedures, provided to the PTO on a set of computer listings, that describe in	0
12	05-20-2016	CRF Disk Has Been Received by Preexam / Group / PCT	0
11	03-23-2016	Application ready for PDX access by participating foreign offices	0
10	03-22-2016	SEQUENCE ERRORS	0
9	03-22-2016	Filing Receipt	0
8	03-22-2016	Applicant Has Filed a Verified Statement of Small Entity Status in Compliance with 37 CFR 1.27	0
7	03-07-2016	PTO/SB/69-Authorize EPO Access to Search Results	0
6	03-07-2016	Applicants have given acceptable permission for participating foreign	0
5	03-10-2016	Cleared by L&R (LARS)	0
4	03-09-2016	Referred to Level 2 (LARS) by OIPE CSR	0

3	03-07-2016	IFW Scan & PACR Auto Security Review	0
2	03-07-2016	Entity Status Set To Undiscounted (Initial Default Setting or Status Change)	0
1	03-07-2016	Initial Exam Team nn	0
0.5	03-07-2016	Filing date	0

Close Window

DRAFT COPY

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT : 10,947,589 B2
DATED : *Mar, 16, 2021
INVENTOR(S): Hicks et al.

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

On the cover page,

[*] Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 45 days.

Delete the phrase “by 45 days” and insert – by 66 days-

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : 10,947,589 B2
APPLICATION NO. : 15/063278
DATED : March 16, 2021
INVENTOR(S) : Hicks et al.

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

On the Title Page:

The first or sole Notice should read --

Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 66 days.

Signed and Sealed this
Seventh Day of February, 2023

Katherine Kelly Vidal

Katherine Kelly Vidal
Director of the United States Patent and Trademark Office

UNITED STATES
PATENT AND TRADEMARK OFFICE

P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

ACKNOWLEDGEMENT OF LOSS OF ENTITLEMENT TO ENTITY STATUS DISCOUNT

APPLICATION #	FILING OR 371(C) DATE	FIRST NAMED APPLICANT	ATTORNEY DOCKET #	REQUEST ID
15/063,278	03/07/2016	James Hicks	81503-AA/GJG/AWG/AC	261594

The entity status change request below filed through Patent Center on 08/16/2024 has been accepted.

Certifications

APPLICANT CHANGING TO REGULAR UNDISCOUNTED FEE STATUS

Signature

I certify, in accordance with 37 CFR 1.4(d)(4), that I am one of the signatories making the entity status change.

Signature	Name	Registration #
/Gary J Gershik/	Gary Gershik	39992