UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE PATENT TRIAL AND APPEAL BOARD

PARSE BIOSCIENCES, INC.
Petitioner,

v.

10X GENOMICS, INC.
Patent Owner

Case No. IPR2023-____
U.S. Patent No. 10,697,013

PETITION FOR *INTER PARTES* REVIEW OF U.S. PATENT NO. 10,697,013

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1. Claim 1: 27

a. [1A] A method for multiplexed analysis of nucleic acids from single cells, the method comprising: 27

b. [1B] providing a sample comprising a plurality of cells, wherein a single cell of the plurality of cells comprises a plurality of sample polynucleotides; 28

c. [1C] performing combinatorial tagging to generate a plurality of tagged polynucleotides from said plurality of sample polynucleotides and a plurality of oligonucleotide tags, ... 29

d. [1D] wherein a tagged polynucleotide of the plurality of tagged polynucleotides is generated by providing an extension product by primer extension using a first oligonucleotide tag and a sample polynucleotide of said plurality of sample polynucleotides, and: 31

e. [1E] ligating a second oligonucleotide tag to said extension product, and 34

(i) The Combination of Linnarsson and McCloskey, and the Knowledge of a POSA Teaches This Limitation 34

f. [1F] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises: a sample sequence corresponding to said sample polynucleotide of the plurality of sample polynucleotides; 39

g. [1G] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises:…a first tag sequence distinguishing said sample polynucleotide from sample polynucleotides from other cells; and..... 42

h. [1H] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises:…a second tag sequence distinguishing said sample polynucleotide from other sample polynucleotides from said cell; 43

i. [1I] amplifying said tagged polynucleotide, thereby generating a plurality of amplified polynucleotides corresponding to the tagged polynucleotide; and..... 44

j. [1J] sequencing said plurality of amplified polynucleotides to determine sequences of the amplified polynucleotides corresponding to the sample sequence, the first tag sequence, and the second tag sequence of the tagged polynucleotide; and..... 44

k. [1K] using the sequences determined in step (d) to count sample polynucleotides for multiple different sample polynucleotides of multiple different single cells of said plurality of cells. 45

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3. Claim 3: The method of claim 2, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to

said mRNA and (ii) extending said first oligonucleotide tag using said mRNA as a template to generate complementary DNA (cDNA). 46

4. Claim 4: The method of claim 3, wherein step (B) comprises (i) ligating said second oligonucleotide tag to said cDNA. 46

5. Claim 5: The method of claim 3, wherein said hybridizing comprises hybridizing a poly-dT sequence of said first oligonucleotide tag to a poly-dA sequence of said mRNA. 47

6. Claim 6: The method of claim 1, wherein said plurality of cells is provided in a plurality of wells. 47

7. Claim 7: The method of claim 1, wherein said first oligonucleotide tag comprises a sequence that is configured to hybridize to said sample polynucleotides 47

8. Claim 8: The method of claim 1, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to said sample polynucleotide and (ii) extending said first oligonucleotide tag or said sample polynucleotide or both to provide said extension product. 47

9. Claim 9: The method of claim 1, wherein step (b) comprises randomly associating said plurality of sample polynucleotides with said plurality of oligonucleotide tags. 47

10. Claim 10: The method of claim 1, wherein said second tag sequences of said plurality of oligonucleotide tags are random sequences. 48

11. Claim 11: The method of claim 1, wherein step (c) comprises generating said plurality of amplified polynucleotides using polymerase chain reaction (PCR). 49

12. Claim 12: The method of claim 1, wherein substantially every sample polynucleotide of said plurality of sample

polynucleotides is associated with the same first tag sequence. 49

13. Claim 19: The method of claim 1, wherein an oligonucleotide tag of said plurality of oligonucleotide tags comprises a sequencing adaptor. 51

14. Claim 20: The method of claim 19, wherein step (d) comprises hybridizing said amplified polynucleotides or derivatives thereof to a solid support via said sequencing adaptor or derivative thereof. 52

15. Claim 21: The method of claim 20, wherein said solid support is a bead. 53

16. Claim 22: 54

    a. [22A] The method of claim 1, wherein step (e) comprises (i) using second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide and/or 54

    b. [22B] The method of claim 1, wherein step (e) comprises…(ii) using sample sequences, first tag sequences, and second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide. 54

17. Claim 23: The method of claim 1, wherein step (e) comprises (i) determining the number of different second tags sequences associated with said sample sequence, thereby estimating the number of sample polynucleotides having said sample sequence from said single cell and/or (ii) using second tag sequences of said plurality of amplified polynucleotides to provide a digital count of said sample polynucleotides. 55

18. Claim 24: The method of claim 1, wherein said plurality of sample polynucleotides are pooled with sample polynucleotides from other single cells of said plurality of cells prior to the generating of step (b).....56

19. Claim 25: The method of claim 1, wherein said plurality of tagged polynucleotides are pooled with tagged polynucleotides from other single cells of said plurality of cells prior to the amplifying of step (c). 57

20. Claim 26: The method of claim 1, wherein said plurality of amplified polynucleotides are pooled with amplified polynucleotides from other single cells of said plurality of cells prior to the sequencing of step (d).....57

21. Claim 27: The method of claim 1, wherein step (e) comprises using first tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the single cell from which the sample sequences are derived based on amplified polynucleotides from the same cell having the same first tag sequence. 58

22. Claim 28: The method of claim 1, wherein step (e) comprises using second tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the sample polynucleotide from which the sample sequences are derived based on amplified polynucleotides from the same sample polynucleotide having the same second tag sequence.....59

B. Ground 2.....60

1. Claim 13: The method of claim 1, wherein at least 90 percent of said plurality of tagged polynucleotides have a second tag sequence that is different from second tag sequences of the other tagged polynucleotides.....60

2. Claim 14: The method of claim 1, wherein substantially every tagged polynucleotide of said plurality of tagged

polynucleotides has a second tag sequence that is different from second tag sequences of the other tagged polynucleotides. 62

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PETITIONER'S EXHIBIT LIST

Exhibit #	Description
EX1001	U.S. Patent No. 10,697,013 ("the '013 patent")
EX1002	Declaration of Gregory Cooper, Ph.D.
EX1003	PCT Patent Publication No. WO 2010/117620 to Linnarsson et al. ("Linnarsson")
EX1004	Megan L McCloskey et al., *Encoding PCR Products with Batch-Stamps and Barcodes*, 45 Biochemical Genetics, 761-767 (2007) ("McCloskey")
EX1005	Prosecution File History of U.S. Patent No. 10,697,013
EX1006	Leslie H. Brail et al., *Gene expression in individual cells: analysis using global single cell reverse transcription polymerase chain reaction (GSC RT-PCR)*, 406 Mutation Research Genomics, 45-54 (1999) ("Brail")
EX1007	Hubert Hug et al., *Measurement of Number of Molecules of a Single mRNA Species in a Complex mRNA Preparation*, 221 J. Theor. Bio., 615-624 (2003) ("Hug")
EX1008	Intentionally left blank.
EX1009	Gerald Schochetman et al., *Polymerase Chain Reaction*, 158 J. of Infectious Diseases 1154-1157 (1988) ("Schochetman")
EX1010	Intentionally left blank.
EX1011	H.C. Yang et al., *A genome-wide study of preferential amplification/hybridization in microarray-based pooled DNA experiments*, 34 Nucleic Acids Research 1-11 (2006) ("Yang")
EX1012	Severine A Degrelle et al., *Amplification biases: possible difference among deviating gene expressions*, 9 BMC Genomics 1-15 (2008) ("Degrelle")
EX1013	Brooks E. Miner et al., *Molecular barcodes detect redundancy and contamination in hairpin-bisulfite PCR*, 32 Nucleic Acids Research 1-4 (2004) ("Miner")

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EX1014	Intentionally left blank.
EX1015	U.S. Patent Provisional Application No. 61/164,759 (filed March 30, 2009) (Sven Linnarsson, applicant) ("Linnarsson Provisional")
EX1016	Defendant 10X Genomic's Opening Brief In Support Of Its Motion To Dismiss, *Becton, Dickinson and Company and Cellular Research, Inc. v. 10X Genomics, Inc.*, C.A. No. 18-1800-RGA (Mar. 1, 2019 D. Del.) (No. 19)
EX1017	Defendant 10X Genomic's Reply Brief In Support Of Its Motion To Dismiss, *Becton, Dickinson and Company and Cellular Research, Inc. v. 10X Genomics, Inc.*, C.A. No. 18-1800-RGA (Apr. 12, 2019 D. Del.) (No. 22)
EX1018	Scheduling Order, *10X Genomics, Inc. and The Board of Trustees of the Leland Stanford Junior University v. Parse Biosciences, Inc.*, C.A. No. 22-1117-JHS (Mar. 2, 2023 D. Del.) (No. 48)
EX1019	Federal Judicial Caseload Statistics (2022), at https://www.uscourts.gov/statistics-reports/analysis-reports/federal-judicial-caseload-statistics
EX1020	Curriculum Vitae of Gregory Cooper
EX1021	Jonas Binladen et al., *The Use of Coded PCR Primers Enables High Throughput Sequencing of Multiple Homolog Amplification Products by 454 Parallel Sequencing*, 2(2): e197 PLoS ONE 1-9 (2007) (Binladen)
EX1022	Intentionally left blank.
EX1023	Motion Success for Stay Pending IPR In Delaware District
EX1024	Hiroto Okayama and Paul Berg, *High-Efficiency Cloning of Full-Length cDNA*, 2 Molecular and Cellular Biology 161-170 (1982) ("Okayama")
EX1025	Bruce Alberts, et al., "Chapter 6 How Cells read the Genome: From DNA To Protein," *Molecular Biology of the Cell*, Garland Science 299-335 (4th ed. 2002)
EX1026	Assignment Record for U.S. Patent 8,298,767

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EX1027	10x Genomics Legal Notice
EX1028	Declaration of Sylvia D. Hall-Ellis, Ph.D.
EX1029	US Patent No. 8,835,358 B2
EX1030	S. Clancy et al., *Translation: DNA to mRNA to Protein*, Nature Education (2008), at https://www.nature.com/scitable/topicpage/translation-dna-to-mrna-to-protein-393/
EX1031	Cole Trapnell, *Defining cell types and states with single-cell genomics*, 25(10) Genome Research 1491-1498 (2015) ("Trapnell")
EX1032	Gerard Brady et al., *Representative in Vitro cDNA Amplification From Individual Hemopoietic Cells and Colonies*, 2 Methods in Molecular and Cellular Biology 17-25 (1990) ("Brady")
EX1033	James Eberwine et al., *Analysis of gene expression in single live neurons*, 89 Proc. Natl. Acad. Sci. 3010-3014 (1992) ("Eberwine")
EX1034	Maria de Fatima Bonaldo et al., *Normalization and Subtraction: Two Approaches to Facilitate Gene Discovery*, 6(9) Genome Research 791-806 (1996) ("Bonaldo")
EX1035	Sydney Brenner et al., *In vitro cloning of complex mixtures of DNA on microbeads: Physical separation of differentially expressed cDNAs*, 97.4 Proceedings of the Nat'l Academy of Sci. 1665-1670 (2000) ("Brenner")
EX1036	David H. Persing, *Polymerase Chain Reaction: Trenches to Benches*, J. of Clinical Microbiology 1281-1285 (1991) ("Persing")
EX1037	Kazuki Kurimoto et al., *An improved single-cell cDNA amplification method for efficient high-density oligonucleotide microarray analysis*, 34(5) Nucleic Acids Research 1-17 (2006) ("Kurimoto")
EX1038	Shigeyuki Esumi et al., *Method for single-cell microarray analysis and application to gene-expression profiling of GABAergic neuron progenitors*, 60 Neuroscience Research 439-451 (2008) ("Esumi")
EX1039	Y. Shibata et al., *Cloning Full-length, Cap-Trapper-Selected cDNAs by Using the Single-Strand Linker Ligation Method*, 30(6) Biotechniques 1250-1254 (2001) ("Shibata")

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EX1040	U.S. Patent Application 2004/0166499
EX1041	Athina Gkazi, *An Overview of Next-Generation Sequencing*, Technology Networks Genomics Research at Figure 2 (March 17, 2021) at https://www.technologynetworks.com/genomics/articles/an-overview-of-next-generation-sequencing-346532
EX1042	Rami Sukarieh, *Relocalization of elF4E by its binding partners upon stress*, Figure 2 (August 2009) at https://www.researchgate.net/publication/41663010_Relocalization_of_eIF4E_by_its_binding_partners_upon_stress
EX1043	Clyde A. Hutchinson, *DNA sequencing: bench to bedside and beyond*, 35(18) Nucleic Acids Research 6227-6237 (2007) ("Hutchinson")
EX1044	Beáta G Vértessy and Judit Tóth, *Keeping Uracil Out of DNA: Physiological Role, Structure and Catalytic Mechanism of dUTPases*, Acc Chem Res. 1-20 (2009) ("Vértessy")
EX1045	Elaine R. Mardis, *Next-Generation DNA Sequencing Methods*, 9 Annual Review Genomics and Human Genetics 387-402 (2008) ("Mardis")
EX1046	Jay Shendure and Hanlee Ji, *Next-Generation DNA Sequencing*, 26(10) Nature Biotechnology 1135-1145 (2008) ("Shendure")
EX1047	Elaine R. Mardis, *The impact of next-generation sequencing technology on genetics*, 24(3) Trends in Genetics 133-141 (2008) ("Mardis 2")
EX1048	Robert A. Holt and Steven J.M. Jones, *The new paradigm of flow cell sequencing*, 18(6) Genome Research 839-846 (2008) ("Holt")
EX1049	National Human Genome Research Institute, *Nucleotide* (March 21, 2023) at https://www.genome.gov/genetics-glossary/Nucleotide
EX1050	National Human Genome Research Institute, *Polymerase Chain Reaction* (March 21, 2023) at https://www.genome.gov/genetics-glossary/Polymerase-Chain-Reaction
EX1051	Plaintiffs, Becton, Dickinson and Company and Cellular Research, Inc.'s First Amended Complaint, *Becton, Dickinson and Company and Cellular Research, Inc. v. 10X Genomics, Inc.*, C.A. No. 18-1800-RGA (Feb 08, 2019 D. Del.) (No. 15)

Exhibit #	Description
EX1052	Exhibits 26 and 27 attached with Plaintiffs' First Amended Complaint, *Becton, Dickinson and Company and Cellular Research, Inc. v. 10X Genomics, Inc.*, C.A. No. 18-1800-RGA (Feb 08, 2019 D. Del.) (No. 15-3)
EX1053	US 9,708,659 B2
EX1054	Georgi K. Marinov et. al, *From single-cell to cell-pool trancriptomes: Stochasticaly in gene expression and RNA splicing*, 24.3 Genome Research 496-510 (2014) ("Marinov")
EX1055	United States Patent Application Publication to McCloskey et al. ("McCloskey II")
EX1056	Boris Hollas and Rainer Schuler, *A Stochastic Approach to Count RNA Molecules Using DNA Sequencing Methods*, 2812 Algorithms in Bioinformatics 55-62 (2003) ("Hollas")
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EX1058	Intentionally left blank.
EX1059	Intentionally left blank.
EX1060	Reinhard Stöger, *Epigenetics and Obesity*, 9(12) Pharmacogenomics 1851-1860 (2008) at https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2652025/ ("Stöger")
EX1061	Diane P. Genereux et. al, *Errors in the Bisulfite Conversion of DNA: Modulating Inappropriate- and Failed-Conversion Frequencies*, 36 Nucleic Acids Research (2008) ("Genereux")
EX1062	Pierre Nicolas et. al., *The Stem Cell Population of the Human Colon Crypt: Analysis via Methylation Patterns*, 3 Plos Computational Biology 0364-0374 (2007) ("Nicolas")
EX1063	"Using Multiplex Identifier (MID) Adaptors for the GS FLX Titanium Chemistry Basic MID Set" Genome Sequencer FLX System, Technnical Bulletin 004-2009, (Apr. 1, 2009) pp. 1-7. http://454.com/downloads/my454/documentation/technical-bulletins/TCB-09004 Using MultiplexIdentifierAdaptorsForTheGSFLXTitaniumSeriesChemistry-BasicMIDSet.pdf.

I. INTRODUCTION

Parse Biosciences, Inc. ("Petitioner") requests *inter partes* review and cancellation of all claims (the "Challenged Claims") of U.S. Patent No. 10,697,013 ("the '013 Patent"), entitled "Methods for Analyzing Nucleic Acids from Single Cells." The '013 Patent is assigned to 10x Genomics, Inc. ("Patent Owner").

The claims of the '013 Patent pertain to methods of analyzing nucleic acid sequences (*i.e.*, polynucleotides) from different single cells by appending two "tags" to the polynucleotides. The first tag allows one to identify the single cell origin of the polynucleotide, and the second tag allows one to distinguish polynucleotides originating from the same single cell. One then sequences or analyzes the tagged DNA.

But such tagging and sequencing methods are not remotely what the '013 Patent characterizes as the alleged invention. Rather, the specification states that the alleged invention is the so-called "reflex method" for rearranging DNA fragments to allow for analyzing longer DNA sequences. *See* EX1001 at Summary Of The Invention. The specification refers to the "reflex method" dozens of times as the key novel idea in the patent. The simplistic labeling methods that appear in the claims, by contrast, are described as nothing but mere prior art techniques. *See, e.g.*, *id*. at 1:38-39 ("We have previously described methods that enable tagging each of a population of fragmented genomes….").

While this alone ought to raise a red flag as to the patentability of the claims of the ’013 Patent, any doubt is put to rest by Patent Owner’s own admissions in a prior litigation against Becton-Dickinson. There, Patent Owner admitted that methods for tagging, detecting, and sequencing DNA are nothing but “generic, routine, well-known laboratory techniques.” EX1016.017. In Patent Owner’s words, “labeling different objects with different labels” is “as old as the scientific method and is familiar to every student in any introductory science class.” *Id.* at .009. According to Patent Owner, “[m]erely apply[ing] this ‘old solution’ of different labels in the particular technological environment of DNA” is nothing new. *Id.* at .016.

Notably, the patents at issue in the Becton-Dickinson case even included claims directed to the very dual-tagging approach claimed in the ’013 Patent wherein a first tag distinguishes among different cells and a second tag distinguishes among different polynucleotides. *See* EX1016.006; EX1051.117. While Patent Owner will now undoubtedly attempt to argue that this dual-tagging approach was novel, it previously grouped it among the ideas that are “as old the scientific method itself” and based on nothing but routine and conventional methods.

As proven herein, Patent Owner’s position to this effect in its prior litigation against Becton-Dickinson was undeniably correct. The claims of the ’013 Patent are obvious in view of: (1) International PCT Patent Application Publication WO

2010/117620 ("Linnarsson") and (2) a peer-reviewed scientific article entitled "Encoding PCR Products with Batch-Stamps and Barcodes" ("McCloskey").

Specifically, the Linnarsson prior art reference discloses virtually every element of the claims of the '013 Patent, including the use of a tag to track which cell a particular nucleic acid molecule comes from. To the extent Linnarsson does not teach a second tag to distinguish among different molecules, this concept is undeniably taught in McCloskey, which proposes a dual-tagging system of precisely the type claimed in the '013 Patent in the context of a system that operates according to the same principles as the Linnarsson system. As documented herein and in the accompanying declaration of Dr. Gregory Cooper, it would have been straightforward and obvious to combine McCloskey's dual-tagging approach with Linnarsson so as to achieve the claims of the '013 Patent.

Accordingly, the Board should institute review of the '013 Patent and find all claims unpatentable.

II. MANDATORY NOTICE

A. Real Party-in-Interest

The real party-in-interest is Parse Biosciences, Inc.

B. Related Matters

As of the filing date of this Petition, the '013 Patent is involved in litigation in *10x Genomics v. Parse Biosciences, Inc.*, 22-cv-01117 (D. Del. filed Aug. 8,

2022). Additionally, in parallel with the instant Petition, Petitioner is seeking review of U.S. Patent Nos. 10,155,981 and 10,240,197 in co-pending *inter partes* reviews.

C. Grounds for Standing

Petitioner certifies that the ’013 Patent is available for *inter partes* review and that Petitioner is not barred or estopped from requesting *inter partes* review challenging the patent claims on the grounds identified in this Petition.

D. Lead and Back-Up Counsel and Service Information

Petitioner provides the following designation of Lead and Back-Up Counsel.

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III. IDENTIFICATION OF CHALLENGES

Petitioner presents the following challenges:

- **Ground 1**: Claims 1-12 and 19-28 of the ’013 Patent are unpatentable under 35 U.S.C. § 103 as being obvious over Linnarsson in view of McCloskey, and further in view of the knowledge of a POSA.
- **Ground 2**: Claims 13-18 of the ’013 Patent are unpatentable under 35 U.S.C. § 103 as being obvious over Linnarsson in view of McCloskey, McCloskey II, and further in view of the knowledge of a POSA.

IV. RELATIONSHIP BETWEEN THIS IPR AND RELATED PROCEEDINGS

In addition to seeking review of the ’013 Patent in the instant petition, Petitioner is also seeking review of U.S. Patent Nos. 10,155,981 (“the ’981 Patent”) and 10,240,197 (“the ’197 Patent”) in co-pending *inter partes* reviews. Petitioner requested review of the ’981 Patent on April 20, 2023. *See* Paper No. 1, IPR2023-00876.

The claims of the ’013 Patent at issue here are highly similar to those of the ’981 and ’197 Patents. Accordingly, the technical background material appearing in the Petition and supporting expert declaration in IPR2023-00876 is directly applicable to not only the instant Petition but also the co-pending petition directed to the ’197 Patent. For the convenience of the Board, Petitioner notes that it should be sufficient for the Board to review only the technical background sections in

IPR2023-00876 to gain an understanding of the necessary technical subject matter relevant to all three petitions.

V. OVERVIEW OF THE ’013 PATENT

A. Priority Date of the ’013 Patent

The ’013 Patent, which was filed on March 12, 2020, identifies two provisional applications, the earlier of which was filed on August 20, 2009. EX1001 at (60). Thus, the earliest priority date of the ’013 Patent is August 20, 2009.

B. The Brenner Family And The Reflex Method

The subject matter of the ’013 Patent was not developed by Patent Owner, but rather by a now-defunct company called PGT in the 2009 time frame. Patent Owner purchased the patent rights from PGT in 2016, and filed the application that resulted in the ’013 Patent in 2020. EX1026; EX1005.098.

The ’013 Patent shares an identical specification with other patents (collectively, “the Brenner Patents”), four of which were filed by PGT and later purchased by Patent Owner in 2016[1]. The Brenner Patents purport to solve a problem with prior art DNA sequencing methods, which were often only able to sequence a short stretch of DNA sequence within much larger fragments of genomic

[1] The PGT Patents include U.S. 8,298,767, 8,563,274, 8,679,756, and 9,102,980. EX1026.005. There is no indication that Patent Owner practices the PGT Patents. *See* EX1027.024-027.

DNA. EX1001 at 1:44-50. As the Brenner Patents state, the "present invention removes the limitations imposed by current sequencing technologies as well as being useful in a number of other nucleic acid analyses." *Id.* at 1:57-59. To this end, the Brenner Patents purport to teach the so-called "reflex method," a technique for breaking up a long sequencing project into a series of smaller individual sequencing projects, the result of which can be pieced together to generate a long stretch of DNA sequence. *See id.* at 17:20-19:21.

The Brenner Patents are clear that this technique of introducing sequences via the "reflex" method is "the invention" itself. *See, e.g., id.* at 12:60-64. The "reflex" method is the very first thing mentioned in the "Summary Of The Invention" section, and it is referred to no less than 225 additional times throughout the common specification of the Brenner Patents. *See generally id.* Every example and embodiment pertains to this "reflex" method.

Rather than pertaining to the "reflex" method, however, Patent Owner's late-drafted claims in the '013 Patent pertain to the concept of putting tags on a DNA sequences so that one can track where the DNA sample originated from. But as explained below, the Brenner Patents characterize this "tagging" process not as an invention, but rather as a prior art technique that the inventors and others had previously accomplished. *See, e.g.*, *id.* at 1:38-41.

C. Overview Of The Claimed Methods And Disclosure Related To "Tagging"

The Challenged Claims are directed to methods for multiplexed analysis of genetic sequences (*i.e.*, polynucleotides) from different single cells by appending "tags" to the polynucleotides. *Id.* at Claim 1; *id.* at Abstract. A full listing of the challenged "tagging" claims appears in the Appendix to this Petition.

Briefly, tagging involves adding synthetic DNA to a sequence of interest by various methods, including ligation or annealing techniques. EX1002 ¶ 71. As acknowledged in the '013 Patent, tagging is not the invention, but was rather a prior art concept. For example, the specification recites: "We have previously described methods that enable tagging each of a population of fragmented genomes." EX1001 at 1:38-41; *see also id.* at 15:28-37 (incorporating by reference prior art references "for their description of nucleic acid tags and their use in identifying polynucleotides"); EX1002 ¶ 71. The '013 Patent acknowledges that "[a]ny convenient method for tagging and/or counting individual nucleic acid molecules with unique tags may be employed." EX1001 at 22:47-49

And indeed, it was common practice to use tags as unique "barcode" identifiers or primers to facilitate amplification, for counting, tracking, or tracing molecules to their sources, and/or for removing amplification bias. *See* EX1002 ¶¶ 72-74; EX1034.003; EX1035.001; EX1007.001; EX1004.003; EX1013.001; EX1003 at 19:4-5. For example, Linnarsson discloses a method for preparing a

cDNA library by tagging cDNA samples originating from different cells with unique tags that identify which single cell each cDNA sample originated from. EX1003 at 19:4-5.

Tagging methods known in the art also involved the use of multiple tags or a combination of tags. EX1002 ¶ 75. For instance, McCloskey discloses a method of analyzing single cell samples by using an encoding oligonucleotide with five information regions, two of which are tag sequences: (1) a "barcode" that uniquely identifies and distinguishes polynucleotides "arising from different cell or allele copies"; and (2) a "batch-stamp" that is "unique to each experiment" and "specifies the DNA source such as the patient or sample identification." EX1004.002, .004. Linnarsson also discloses tagging a cDNA sample with a combination of tags. EX1003 at 19:6-7. It was also routine and conventional to use a large number of tags to ensure efficient tagging of all polynucleotides in a cell. EX1002 ¶ 76; EX.1056.001.

The claims of the '013 Patent recite the prior art approach of using two tags to count and track through amplification and sequencing two types of information available in the originating sample. The tags allow a researcher to distinguish (1) from which single cell the polynucleotide originates, and (2) one polynucleotide from another within that single cell.

D. Prosecution History Overview

During prosecution, the McCloskey reference was not before the Examiner. As to Linnarsson and McCloskey II, the Applicant submitted an Information Disclosure Statement identifying these references, *see* EX1005.005, 007, 142, 144, but the Examiner otherwise did not cite or discuss either Linnarsson or McCloskey II during prosecution.

In fact, the Examiner did not issue any rejections whatsoever during prosecution of the ’013 Patent, and allowed all claims after the Applicant filed a terminal disclaimer disclaiming the terminal portion of the statutory term of the ’013 Patent that would extend beyond the expiration dates of U.S. 10,155,981 (the “’981 Patent”), U.S. 10,240,197 (the “’197 Patent”), U.S. 10,280,459, U.S. 10,337,063, U.S. 10,392,662 and Application No. 16/443,703. EX1005.130.

In the Reasons for Allowance, the Examiner stated that the prior art which was before the Examiner did not teach the claimed method of combinatorial tagging of polynucleotides from single cells with two tag sequences:

> The claims are allowed for the reason that…prior art does not teach or fairly suggest Applicants’ claimed method of combinatorial tagging polynucleotides from single cells in which a sample polynucleotide of a plurality of sample polynucleotides comprises a first tag sequence which distinguishes the sample polynucleotide from the sample polynucleotides from other cells and a second tag

> sequence which distinguishes the sample polynucleotide from other sample polynucleotides.

EX1005.131. McCloskey, which was not before or expressly considered by the Examiner, however, discloses precisely this method of tagging with two tag sequences. Specifically, McCloskey discloses using a first tag to distinguish polynucleotides from different cells and a second tag to distinguish one polynucleotide from other polynucleotides from the same single cell. *See infra* Sections IX.A.1.e-h. Thus, the Examiner's decision to allow the claims was based solely on an incomplete and wholly inadequate understanding of the scope of the prior art.

E. Patent Owner's Admissions Regarding The Prior Art And The Novelty Of The Claimed Inventions

Patent Owner's prior admissions confirm unequivocally that the claims of the '013 Patent are not remotely patentable and that the '013 Patent issued only because the Examiner erred in its analysis of the prior art during prosecution.

In an unrelated case between Patent Owner and Becton, Dickinson and Company ("Becton-Dickinson"),[2] Patent Owner challenged the eligibility of seven asserted patents (the "Fodor Patents"), which are directed to tagging DNA molecules

[2] *Becton, Dickinson and Company et al. v. 10X Genomics, Inc.*, C.A. No. 18-1800-RGA (D. Del.).

for the purpose of tracking and counting individual molecules. *See* EX1016.007-008. This is the exact thing to which the claims of the '013 Patent are directed. Petitioner's expert, Dr. Gregory Cooper, explains in detail the similarity between the claims at issue in the prior Becton-Dickinson case and the claims at issue now. *See* EX1002 ¶¶ 38-42.

According to Patent Owner, the Fodor Patents are "nakedly directed at the abstract idea of labeling different objects (two or more 'nucleic acid molecules', e.g., portions of DNA) with different labels ('a plurality of nucleic acid label-tags with different sequences.')." EX1016.010. Patent Owner argued that the concept of "labeling different objects with different labels" is "as old as the scientific method and is familiar to every student in any introductory science class." *Id.* at .009. And the Fodor Patents "[m]erely apply[] this 'old solution' of different labels in the particular technological environment of DNA." *Id.* at .016.

Patent Owner further argued that elements of the Fodor Patents—which also are claimed by the '013 Patent—are "***well-known and conventional***." *Id.* at .011. In Patent Owner's words, the method for attaching a tag to a DNA sample was "well-known and conventional before 2009." *Id.*; *compare with* EX1001 at Claim 1 ("(b) generating a plurality of tagged polynucleotides").

Indeed, one of the concepts that Patent Owner characterized as being "as old as the scientific method" is the very idea of dual-tagging, as claimed by claims 13

and 19 of one of the Fodor Patents (U.S. Pat. 9,708,659). *See* EX1053 at claims 13, 19; EX1051.117 (Becton-Dickinson's complaint asserting claims 13 and 19 of the '659 Patent); EX1052.013-019, .031-039 (Becton-Dickinson's infringement charts for these claims accusing 10X's dual-tag single-cell analysis system). As explained by Dr. Cooper, these two Fodor claims are directed to tagging all molecules in a cell with the same cell tag, and further tagging individual molecules within the cell with an additional molecular identifier. EX1002 ¶ 42 (explaining that these claims are directed to the idea of dual tagging). Patent Owner challenged these claims, among others, as being well-known.

Patent Owner also confirmed that detecting and sequencing DNA samples were nothing but "generic, routine, well-known laboratory techniques." EX1016.018; *compare with* EX1001 at Claim 1 ("(d) sequencing said plurality of amplified polynucleotides"). As shown in the chart that ***Patent Owner*** submitted in its prior litigation against Becton-Dickinson, Patent Owner admitted that elements common to both the Fodor Patents (*e.g.*, claim 6 of U.S. Patent No. 8,835,358) and the '013 Patent are "[g]eneric, conventional [] techniques." EX1016.010-011.

358 patent Claim 6	Summary
6. A method comprising:	Preamble
(a) combining a mixture comprising at least two distinct target nucleic acid molecules with a pool of nucleic acid label-tags, wherein the pool of nucleic acid label-tags comprises a plurality of nucleic acid label-tags with different sequences;	Different molecules ("two distinct target nucleic acid molecules") and Different labels ("a pool of nucleic acid label-tags")
(b) attaching at least two nucleic acid label-tags from the pool of nucleic acid label-tags to the at least two distinct target nucleic acid molecules to obtain at least two label-tag-target nucleic acid molecules, wherein the distinct target nucleic acid molecules have different sequences from one another;	Generic, conventional attaching techniques
(c) amplifying at least a portion of the label-tag-target nucleic acid molecules, wherein an amplified portion of the label-tag-target nucleic acid molecules comprises at least a portion of said target nucleic acid molecule; and	Generic, conventional amplifying techniques
(d) detecting an amplified product of step (c).	Generic, conventional detecting techniques

Id. at .011. Notably, as shown above, Patent Owner even said that the idea of "attaching at least two nucleic acid label tags" to a DNA molecule reflects nothing but "[g]eneric, conventional attaching techniques." This is the very thing that Patent Owner now relies upon as the point of novelty in the '013 Patent.

VI. LEVEL OF ORDINARY SKILL IN THE ART OF THE '013 PATENT

A person of ordinary skill in the art ("POSA") relating to the '013 Patent would have had (1) a Masters degree and/or Ph.D. in molecular biology, genetics, chemistry, engineering, or a related discipline and at least two years of post-Masters, postdoctoral, or industry experience or (2) a Bachelor of Science in such disciplines and at least five years of academic or industry experience (including any experimental work toward a graduate degree), relating to DNA sequencing technologies, amplification methods, and gene expression analysis. EX1002 ¶ 51. Further, a POSA would have been familiar with associated tools, methods, and

techniques including: (1) polynucleotide tagging, (2) amplification, (3) use of tags to overcome amplification bias, (4) single cell analysis, (5) ligation and primer extension, and (6) sequencing. EX1002 ¶¶ 52, 74-95.

VII. THE CITED PRIOR ART

A. Linnarsson

Linnarsson, entitled "Gene Expression Analysis In Single Cells," was published on October 14, 2010 as international PCT patent application publication WO 2010/117620 A2. EX1003.001. Linnarsson claims priority to U.S. Provisional Patent Application No. 61/164,759 ("Linnarsson Provisional"), filed on March 30, 2009. EX1015 at 1:3-5.

1. Linnarsson Is § 102(e) Prior Art

Linnarsson is § 102(e) prior art as of at least March 30, 2009, the date of the filing of the Linnarsson Provisional, because the Linnarsson Provisional provides written description support for Linnarsson's claimed invention of preparing a cDNA library by tagging cDNAs with unique identifiers. *See* MPEP § 2136; *Dynamic Drinkware, LLC. v. Nat'l Graphics, Inc.*, 800 F.3d 1375, 1382 (Fed. Cir. 2015); *In re Giacomini*, 612 F.3d 1380, 1383 (Fed. Cir. 2010)

Indeed, Linnarsson was cited as prior art during prosecution of the '013 Patent, and the Applicants did not contest its prior art status. Instead, the Applicants identified Linnarsson as prior art in an Information Disclosure Statement ("IDS").

EX1005.125. Therefore, Linnarsson is prior art to the Challenged Claims even if the Challenged Claims are entitled to their earliest priority date of August 20, 2009.

The following chart shows that each and every limitation of claim 1 of Linnarsson is disclosed by the Linnarsson Provisional Application:

Linnarsson	**Linnarsson Provisional Application Disclosure**
1. A method of preparing a cDNA library from a plurality of single cells, the method comprising the steps of:	"In particular, the invention relates to a method for preparing a cDNA library from a plurality of single cells, and to a cDNA library produced by this method." EX1015 at 1:4-6; *see also id.* at 2:23-25, 8:33-35.
(i) releasing mRNA from each single cell to provide a plurality of individual mRNA samples, wherein the mRNA in each individual mRNA sample is from a single cell;	"the method comprising: (i) releasing mRNA from each single cell to provide a plurality of individual samples, wherein the mRNA in each individual mRNA sample is from a single cell;" EX1015 at 2:28-32; *see also id.* at 3:5-6, 4:12-15.
(ii) synthesizing a first strand of cDNA from the mRNA in each individual mRNA sample and incorporating a tag into the cDNA to provide a plurality of tagged cDNA samples, wherein the cDNA in each tagged cDNA sample is complementary to mRNA from a single cell;	"(ii) synthesising a first strand of cDNA from the mRNA in each individual mRNA sample and incorporating a tag into the cDNA to provide a plurality of tagged cDNA samples, wherein the cDNA in each tagged cDNA sample is complementary to mRNA from a single cell;" EX1015 at 2:33-37; *see also id.* at 5:4-7, 5:24-32.
(iii) pooling the tagged cDNA samples; and	"pooling the tagged cDNA samples; and" EX1015 at 3:1; *see also id.* at 3:9-20.

Linnarsson	Linnarsson Provisional Application Disclosure
(iv) amplifying the pooled cDNA samples to generate a cDNA library comprising double-stranded cDNA,	"amplifying the pooled cDNA samples to generate a cDNA library comprising double-stranded cDNA." EX1015 at 3:2-3; *see also id.* at 11:14-17, 14:5-7.

Thus, Linnarsson is § 102(e) prior art, because at least one of its published claims is supported by the disclosure of the Linnarsson Provisional Application, which was filed before the earliest possible priority date of the '013 Patent. *See supra Dynamic Drinkware*; *In re Giacomini*. Moreover, the disclosures of Linnarsson that Petitioner relies upon for the ground are supported by the disclosure of the Linnarsson Provisional Application, as shown below by parallel citations.

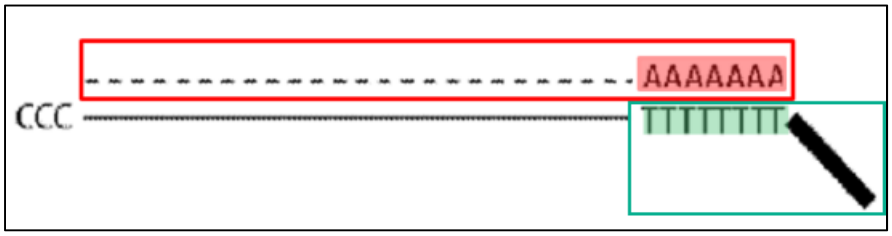
2. Linnarsson Overview

Linnarsson is directed to analyzing gene expression of a plurality of single cells. EX1003 at 1:6. In particular, Linnarsson discloses a method of preparing a cDNA library for sequencing by appending unique oligonucleotide sequences, or "tags," to cDNA samples originating from a plurality of cells. *Id.* at 10:15-22. Linnarsson teaches that its method is useful for distinguishing polynucleotides arising from different single cells in a multi-cell experiment, thus reducing cell-to-

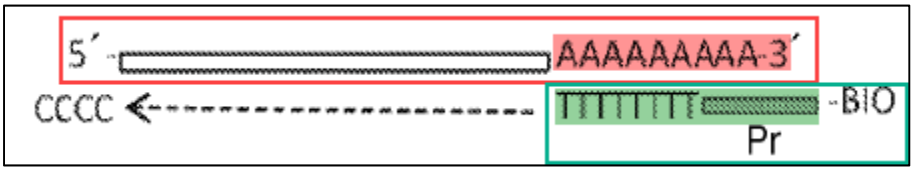
cell amplification bias. *Id.* at 37:28-29, 19:9-10 (disclosing that the tag acts as a barcode to "identify which single cell from each cDNA sample originated.").

According to Linnarsson's method, tagged cDNA is synthesized from mRNA by reverse transcription using a tagged cDNA synthesis primer ("CDS") and/or a tagged template switching oligonucleotide ("TSO") to incorporate a tag into the resulting cDNA. EX1003 at 18:31-32 ("In some aspects of the invention, a tag can be incorporated into the cDNA during its synthesis. For example, the CDS and/or the TSO can include a tag."). Linnarsson further discloses performing PCR on the tagged cDNA and subsequently analyzing and sequencing the tagged cDNA. EX1003 at 3:29-4:2, 19:16-17, 21:13-20.

Specifically, Linnarsson discloses a cDNA synthesis primer ("CDS") (boxed in green) that includes a sequence of thymines (highlighted in green) and that "directs synthesis of a cDNA strand" by hybridizing with a sequence of adenine nucleotides making up the poly-A tail (highlighted in red) of an mRNA strand (boxed in red). This is shown in both Figures 2 and 11 of Linnarsson:

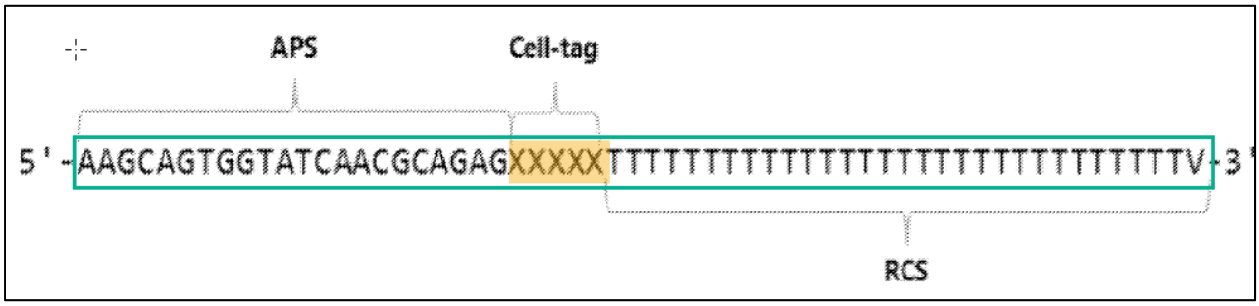


EX1003 at Fig. 2.



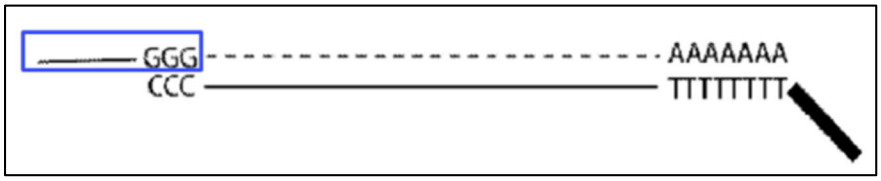
Id. at Fig. 11; *id*. at 5:29-31, 24:13-15; EX1002 ¶ 101. The resulting cDNA molecule also includes at its end a series of cytosine (C) nucleotides, which is added by the natural activity of the enzyme that is responsible for synthesizing the cDNA. EX1003 at 16:22-26.

Figure 4 of Linnarsson depicts an example of a CDS (boxed in green) with a cell-tag (highlighted in yellow):

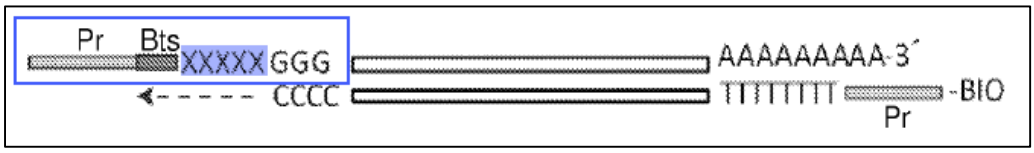


EX1003 at Fig. 4. Linnarsson discloses that the CDS cell-tag "can be incorporated into the cDNA" and can "be used to identify which single cell from each cDNA sample originated." EX1003 at 18:31, 19:9-10.

Linnarsson also discloses a template switching oligonucleotide ("TSO") (boxed in blue) that comprises the tag "XXXXX" (highlighted in blue), which "anneals transiently" to the series of cytosine nucleotides at the end of the newly-synthesized strand "and cDNA synthesis proceeds using the oligo as template." This too is shown in both Figures 2 and 11 of Linnarsson:

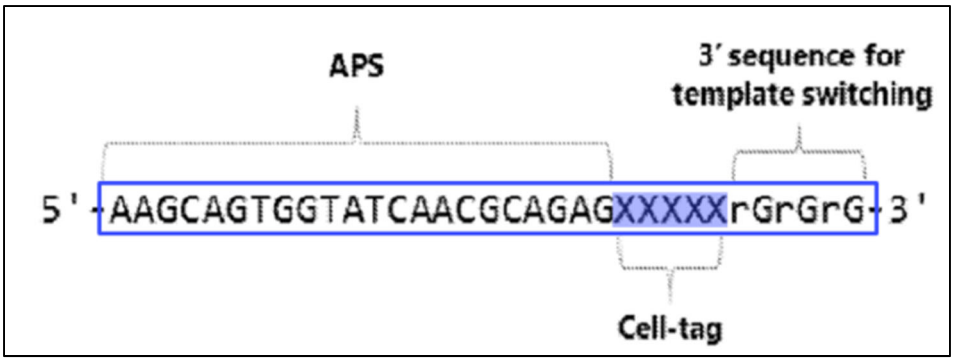


EX1003 at Fig. 2.



Id. at Fig. 11; *id.* at 5:31-33, 24:15-20 (explaining Figure 11); EX1002 ¶¶ 113-114.

Figure 3 similarly depicts an example of a TSO (boxed in blue) with a cell-tag (highlighted in blue):



EX1003 at Fig. 3. Linnarsson discloses that the TSO cell-tag "can be incorporated into the cDNA" and can "be used to identify which single cell from each cDNA sample originated." *Id.* at 18:31, 19:9-10.

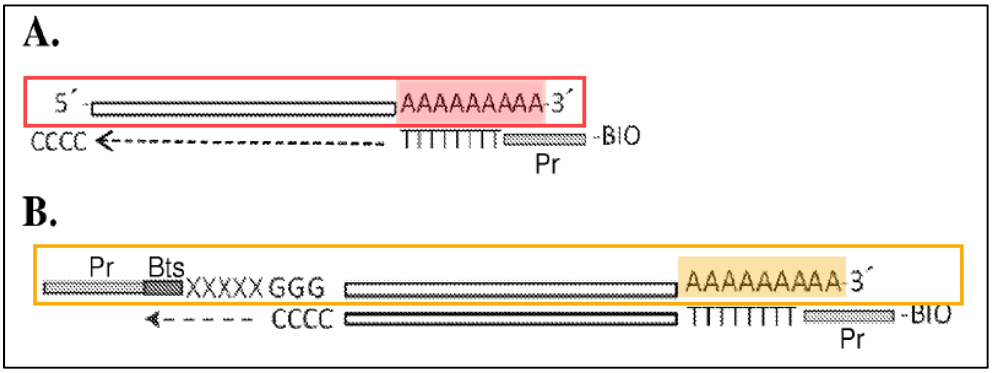
Thus, Linnarsson discloses that the tagged CDS and tagged TSO can be used alone or in combination to identify the single-cell origin of the tagged cDNA. EX1003 at 18:32 (The "CDS and/or the TSO can include a tag."); *id.* at 19:4-5 ("As

the tag is present in the CDS and/or the TSO it will be incorporated into the cDNA during its synthesis and can therefore act as a 'barcode' to identify the cDNA.").

Linnarsson further teaches that the tagged cDNA can be amplified by PCR. EX1003 at 19:16-17. Linnarsson explains that "PCR [and] ligation amplification (or ligase chain reaction, LCR)…are known and widely practiced in the art." EX1003 at 12:18-20. Linnarsson also teaches that after generating the tagged cDNA, "sequencing is performed" thereon. *See* EX1003 at 4:8-13, 19:28-32, 21:13-25 ("This cDNA library can be sequenced to provide an analysis of gene expression in single cells or in a plurality of single cells.").

Notably, during reverse transcription, the sample mRNA is denatured from the first synthesized cDNA strand and a new complementary strand of cDNA is formed along the first synthesized cDNA strand, generating a double-stranded cDNA. As a result, one strand of the double-stranded cDNA molecule (boxed in orange) has the same sequence as the original mRNA strand (boxed in red), including the same sequence of adenines (highlighted in orange) that made up the poly-A tail (highlighted in red) of the mRNA:

EX1003 at Fig. 2. Figure 11 of Linnarsson discloses the same:



Id. at Fig. 11; *id.* at 6:1-3 (explaining Figure 11); *see also* EX1002 ¶ 108.

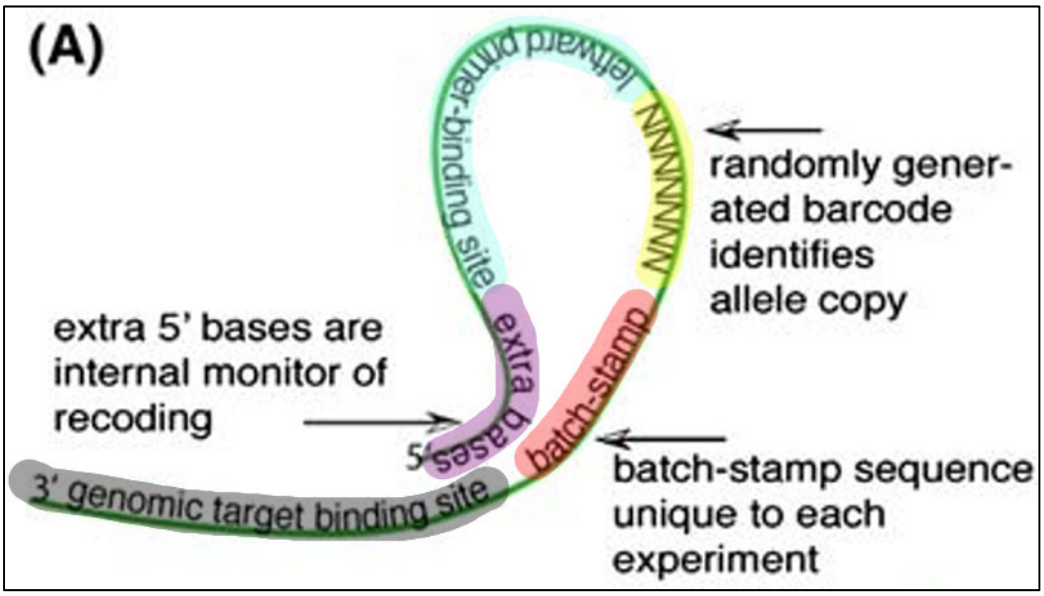
B. McCloskey

The McCloskey reference, titled "Encoding PCR Products with Batch-Stamps and Barcodes," is a peer-reviewed scientific article published in the December 2007 print issue of the Biochemical Genetics journal. The journal published the McCloskey reference online on October 23, 2007. EX1028; EX1004.001. McCloskey is prior art under pre-AIA 35 U.S.C. § 102(a) and (b). The Examiner did not consider McCloskey during prosecution.

McCloskey discloses a method of DNA analysis for single-cell samples that utilizes an "encoding oligonucleotide" having "five distinct information regions,"

two of which are tag sequences that distinguish, respectively, polynucleotides and samples.

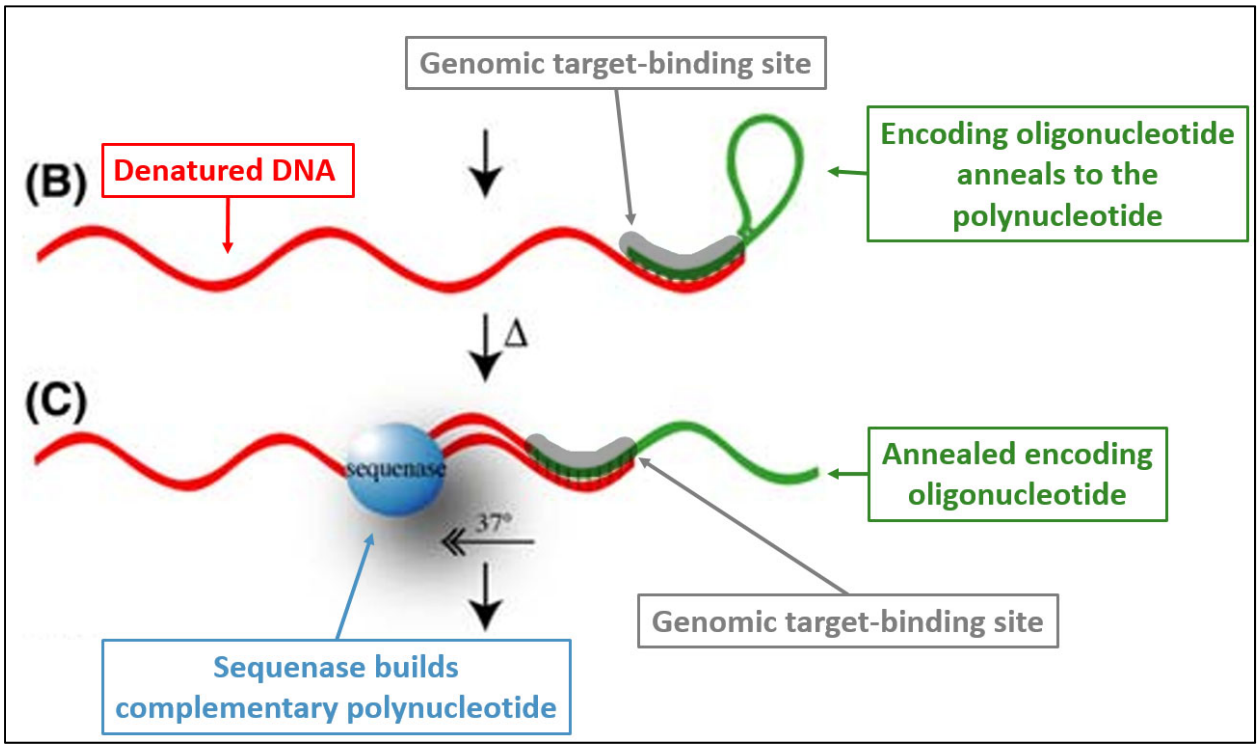
As depicted below, McCloskey teaches a first tag sequence, the "barcode" (highlighted in yellow), which "distinguishes among sequences arising from different cell or allele copies." EX1004.002-004. McCloskey further teaches a second tag sequence, the "batch-stamp" (highlighted in red), that is unique to each experiment and is intended to associate a polynucleotide to "a specific sample run on a specific date." *Id*. at .004.



Id. at Fig. 1.

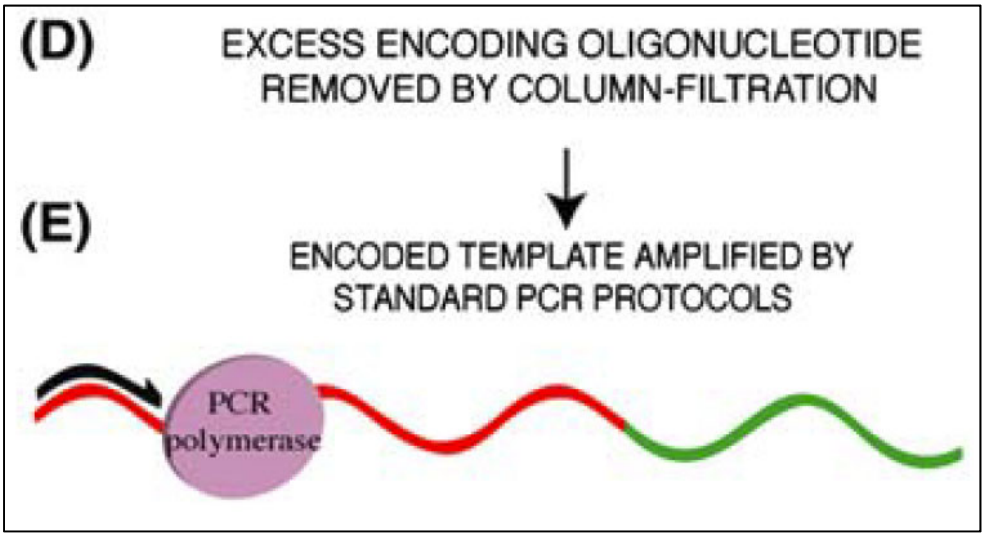
According to McCloskey's method, the encoding oligonucleotide's genomic target binding site (shown in grey, above) anneals to a denatured polynucleotide strand and is then extended by Sequenase, a type of DNA polymerase. *Id.* at .002-

005. The annealing and extension steps are shown at the top and bottom, respectively, of the figure below:



Id. at Fig. 1.

After the annealing and extension steps, McCloskey teaches purifying the sample to remove encoding oligonucleotides that were not annealed, followed by PCR amplification and sequencing. *Id.* at .005-007 ("Sequenase extension products were purified to remove any unincorporated encoding oligonucleotides using the Qiagen QIAquick PCR purification kit."). These steps are illustrated below:



Id. at Fig. 1.

McCloskey teaches that an advantage of its method is that it allows an experimenter to distinguish redundant sequences that are the result of PCR amplification from redundant sequences that existed in the sample before amplification. Two identical sequences that have the same barcode suggest that the redundancy is from PCR amplification, whereas two identical sequences that have a different barcode suggest that the redundancy existed in the sample prior to amplification. *See id.* at .006 ("Redundant sequences recovered from the same PCR reaction are readily identified because they are identical in both barcode and batch-stamp."). Therefore, the number of unique barcodes identified at the sequencing stage represents the template diversity of the analyzed sample. *Id.* ("An additional benefit of our method for mosaic samples is that counting sequences with nonredundant barcodes provides a quantitative measure of template diversity.").

McCloskey teaches that its method increases reliability: "Our approach thus enables classification of each PCR-derived sequence as valid, contaminant, or redundant, and provides a measure of sequence diversity." EX1004.001; *see also id.* at Figs. 1(F) – 1(I) (showing various barcode and batch-stamp sequences from one experiment), *id.* at .003. McCloskey also recommends "that batch-stamps and barcodes be used when amplifying irreplaceable DNAs and cDNAs for forensic, clinical, ***single cell***, and ancient DNA analyses." *Id*. at .005.

C. McCloskey II

U.S. Patent Publication 2007/0020640 ("McCloskey II"), entitled "Molecular Encoding Of Nucliec Acid Templates For PCR And Other Forms Of Sequence Analysis," was published on January 25, 2007. EX1055.001. McCloskey II is prior art under pre-AIA 35 U.S.C. § 102(a) and (b). During prosecution of the '013 Patent, the Applicant identified McCloskey II as prior art in an IDS. EX1005.005, 142. The Examiner raised no prior art rejection based on McCloskey II.

McCloskey II is a sister patent application that goes along with the McCloskey reference. The subject matter of McCloskey II was invented by the authors of McCloskey. Thus, similar to McCloskey, McCloskey II also discloses a method for tagging polynucleotides with "a molecular barcode and batch-stamp." EX1055 at Abstract. In addition, McCloskey II discloses that its barcode sequence may comprise up to 30 nucleotides. EX1055 at [0021].

VIII. CLAIM CONSTRUCTION

For purposes of this Petition, Petitioner does not believe that any terms require explicit construction in view of the prior art being presented. To the extent Patent Owner identifies terms for construction, Petitioner reserves the right to respond. Claim terms are given their ordinary and customary meaning as interpreted by a POSA at the time of the invention. That being said, Petitioner notes that Claim 1 of the '013 Patent recites "providing a sample." The specification defines a "sample" to mean "a quantity of material from a biological, environmental, medical, or patient source in which detection, measurement, or labeling of target nucleic acids is sought." EX1001 at 12:15-18.

IX. IDENTIFICATION OF HOW THE CHALLENGED CLAIMS ARE UNPATENTABLE

A. Ground 1

As explained below, Claims 1-12 and 19-28 of the '013 Patent would have been obvious to a POSA over the combination of Linnarsson, McCloskey, and the knowledge of a POSA.

1. Claim 1:

a. [1A] A method for multiplexed analysis of nucleic acids from single cells, the method comprising:

To the extent the preamble is limiting, Linnarsson discloses this limitation.

Linnarsson discloses a method for multiplexed analysis of nucleic acids from single cells: the "present invention provides methods and compositions for the

analysis of gene expression in single cells or in a plurality of single cells." EX1003 at 9:23-25; EX1015 at 1:3-8; *see also* EX1003 at Abstract, 10:15-22, 22:22-28; EX1015 at 1:3-8, 2:27-3:3, 9:8-10, 11:8-17. In one embodiment, Linnarsson's method comprises the following steps: "(A) the tissue of interest is dissected; (B) ***a plurality of single cells are selected***; (C) single cells are placed in separate wells of a 96-well plate and lysed; tagged reverse transcription is performed on each sample to produce cDNA; (D) cDNA samples are pooled and amplified; (E) ***sequencing is performed*** to obtain 100 million reads; and (F) ***identification of expressed genes and identification of cells from which they originated***." EX1003 at 4:8-13; EX1015 at 9:35-10:6; *see also* EX1003 at Fig. 1; EX1015 at Fig. 1; EX1002 ¶ 117.

b. [1B] providing a sample comprising a plurality of cells, wherein a single cell of the plurality of cells comprises a plurality of sample polynucleotides;

Linnarsson discloses this claim element.

First, Linnarsson teaches providing a sample comprising a plurality of single cells. EX1002 ¶ 120. According to Linnarsson, this sample of "[c]ells [can be] obtained from a tissue of interest…." EX1003 at 22:22-23; EX1015 at 11:9; *see also* EX1003 at 4:8-10; EX1015 at 9:35-37. Linnarsson explains that the sample of single cells can take various forms:

> Single cells useful in the methods described herein can be obtained from a tissue of interest, or from a biopsy, blood sample, or cell culture.

EX1003 at 13:11-12; EX1015 at 3:31-34. Linnarsson further teaches that these single cells are placed in a 96-well plate for subsequent processing and analysis. *See* EX1003 at 4:10-11, 22:23-24; EX1015 at 9:37-10:1, 11:10-11.

Second, Linnarsson teaches that each single cell of the plurality of single cells comprises a plurality of sample polynucleotides. EX1002 ¶ 124. For example, Linnarsson discloses that the single cells comprise mRNA, which is a polynucleotide. *See* EX1003 at 4:10-11, 13:28; EX1015 at 4:12, 9:37-10:3; *see also* EX1003 at 22:24-26; EX1015 at 11:11-14. Linnarsson further teaches that cells contain a plurality of mRNA molecules. *See, e.g.*, EX1003 at 3:26-30; EX1015 at 2:27-3:3.

c. [1C] performing combinatorial tagging to generate a plurality of tagged polynucleotides from said plurality of sample polynucleotides and a plurality of oligonucleotide tags,

As discussed above in Section V.E, Patent Owner previously alleged that claims directed to a method for attaching a tag to a DNA sample reflected techniques that were "well-known and conventional before 2009." EX1016.010. And indeed, "combinatorial tagging" as recited in this claim element would have been obvious in view of the combination of Linnarsson and McCloskey.

As explained above in Section IX.A.1.b., Linnarsson teaches a plurality of single cells comprising a plurality of sample polynucleotides (*i.e.*, mRNA).

According to Linnarsson, these sample mRNA are released from the single cells via lysis. *See* EX1003 at 4:10-11, 13:28; EX1015 at 4:12, 9:37-10:1.

Linnarsson then teaches performing reverse transcription on the plurality of sample mRNA molecules to synthesize, or generate, a plurality of cDNA molecules. *See* EX1003 at 3:25-30, 22:24-26; EX1015 at 2:27-3:3, 11:11-14; *see also* EX1003 at 14:9-32 (describing reverse transcription methods to synthesize cDNA); EX1015 at 4:29-5:22; EX1002 at ¶ 129.

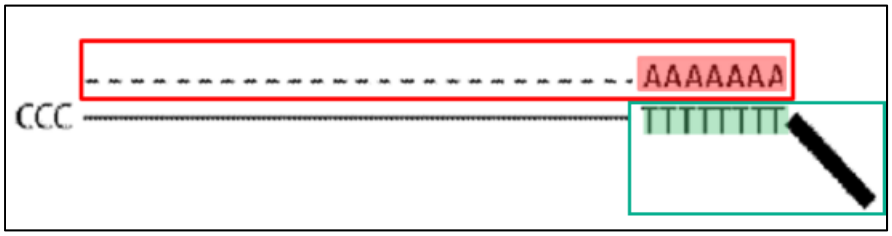
Furthermore, Linnarsson teaches that during cDNA synthesis, oligonucleotide tags (*i.e.*, CDSs and/or TSOs) can be used to tag the cDNA. EX1003 at 19:4-5 ("As the tag is present in the CDS and/or the TSO it will be incorporated into the cDNA during its synthesis and can therefore act as a 'barcode' to identify the cDNA."); EX1015 at 7:23-26; EX1002 ¶ 130. As explained further in Sections IX.A.1.d and IX.A.1.e below, "combinatorial tagging," wherein "one [tag] is attached by ligation and a second [tag] is attached by primer extension" would be obvious in view of Linnarsson and McCloskey. EX1001 at 7:25-27. The double-stranded cDNA molecules that would result from such a process are the claimed "plurality of tagged polynucleotides."

d. [1D] wherein a tagged polynucleotide of the plurality of tagged polynucleotides is generated by providing an extension product by primer extension using a first oligonucleotide tag and a sample polynucleotide of said plurality of sample polynucleotides, and:

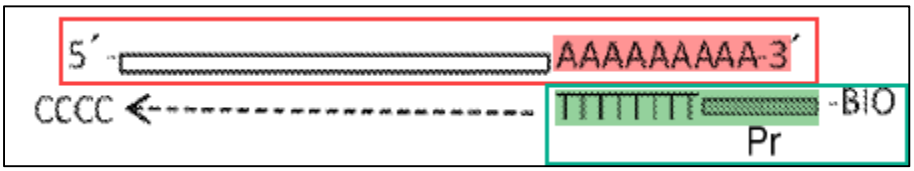
Linnarsson teaches performing reverse transcription, which is a type of primer extension, on the plurality of sample mRNA molecules to generate a plurality of tagged cDNA molecules. *See* EX1003 at 3:25-30, 22:24-26; EX1015 at 2:27-3:3, 11:11-14; *see also* EX1003 at 14:9-32; EX1015 at 4:29-5:22.

Furthermore, Linnarsson teaches tagging the cDNA with a first oligonucleotide tag (*i.e.*, CDSs and/or TSOs) during primer extension. EX1003 at 19:4-7 ("As the tag is present in the CDS and/or the TSO it will be incorporated into the cDNA during its synthesis and can therefore act as a 'barcode' to identify the cDNA."); EX1015 at 7:23-26. The resulting tagged double-stranded cDNA are the claimed "plurality of tagged polynucleotides."

First, Linnarsson teaches using a tagged CDS during primer extension cDNA synthesis to generate tagged cDNA. As shown in Figures 2 and 11, a CDS (boxed in green) is used to "direct[] synthesis of a cDNA strand." The CDS includes a sequence of thymines (highlighted in green) that hybridize with a sequence of adenines making up a poly-A tail (highlighted in red) of an mRNA molecule (boxed in red):

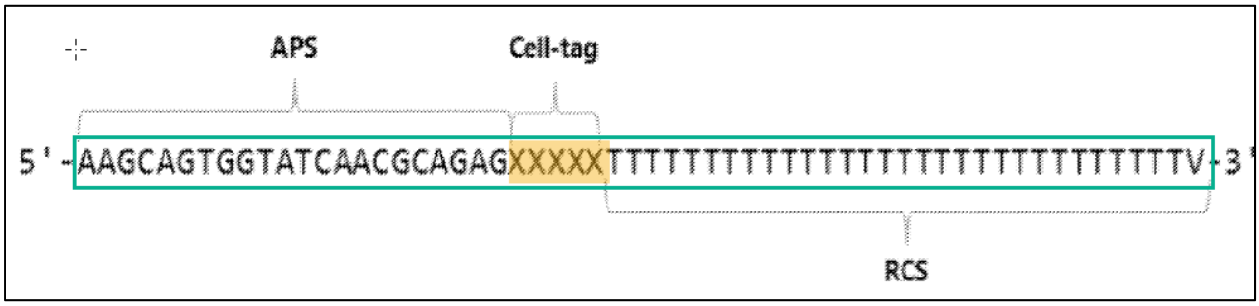


EX1003 at Fig. 2; *see also* EX1015 at Fig. 2. Figure 11 of Linnarsson discloses the same:



Id. at Fig. 11; *see also* EX1015 at Fig. 2; EX 1003 at 5:29-31 (explaining the disclosure of Figure 11); EX1015 at 5:24-32, 13:8-14 (same); EX1002 ¶ 135.

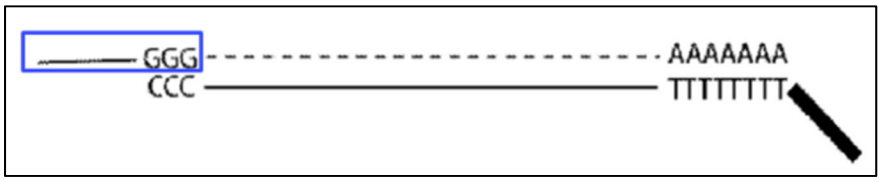
As shown in Figure 4, Linnarsson discloses that the CDS (boxed in green) comprises a cell-tag (highlighted in yellow):



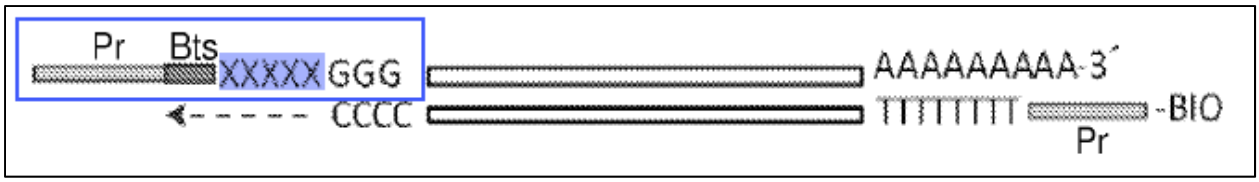
EX1003 at Fig. 4; *see also* EX1015 at Fig. 4.

Second, Linnarsson teaches using a tagged TSO during primer extension to generate tagged cDNA. As shown in Figure 2 below, a TSO (boxed in blue) that comprises the tag "XXXXX" (highlighted in blue) "anneals transiently, and cDNA synthesis proceeds using the oligo as template." EX1003 at 5:31-33; *see also*

EX1015 at 13:14-25; EX1003 at 16:10-17 (discussing another method of forming the second strand of cDNA); EX1015 at 6:1-11; EX1002 ¶¶ 139-140.

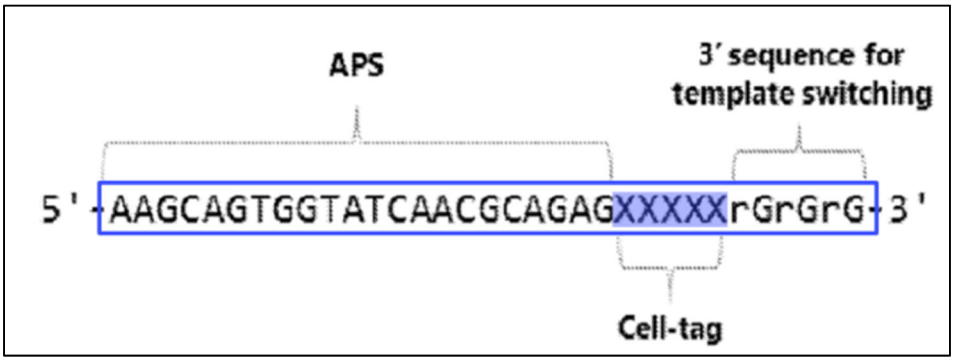


EX1003 at Fig. 2; *see also* EX1015 at Fig. 2. Linnarsson Figure 11 discloses the same:



EX1003 at Fig. 11; EX1015 at Figs. 2, 3.

Figure 3 of Linnarsson similarly discloses a TSO (boxed in blue) comprising a cell-tag (highlighted in blue):



EX1003 at Fig. 3; EX1015 at Fig. 3.

Linnarsson further teaches that "[b]oth the CDS and the TSO can include a tag. The CDS and the TSO can each include a different tag, such that the tagged

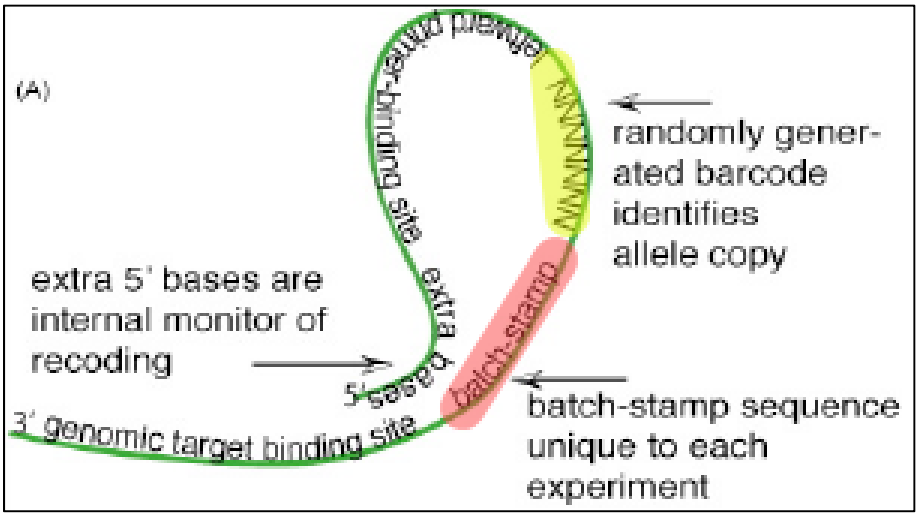
cDNA sample comprises a combination of tags." EX1003 at 19:5-7; EX1015 at 7:23-28.

Thus, Linnarsson teaches providing an extension product by primer extension (reverse transcription) using a first oligonucleotide tag (*i.e.*, TSO and/or CDS) and a sample polynucleotide from the plurality of sample polynucleotides (*i.e.*, mRNA) as required by this claim limitation.

e. [1E] ligating a second oligonucleotide tag to said extension product, and

(i) The Combination of Linnarsson and McCloskey, and the Knowledge of a POSA Teaches This Limitation

To the extent Linnarsson does not teach ligating a second oligonucleotide tag to the claimed "extension product," this would have been obvious in view of McCloskey and the knowledge of a POSA. By way of background, McCloskey teaches an "encoding oligonucleotide" as depicted below in Figure 1:



EX1004 at Fig. 1. As shown above, McCloskey's encoding oligonucleotide comprises *inter alia* two tag sequences: (1) a "batch-stamp" sequence (highlighted in red, above) to identify the experimental batch or genomic source (*e.g.*, a single cell) and (2) a "barcode" (highlighted in yellow, above) to distinguish polynucleotide sequences "arising from different cell or allele copies." *See* EX1004.001-003. As documented below, it would have been obvious to utilize Linnarsson's cDNA library preparation to introduce a cell tag and then further introduce a second tag to distinguish polynucleotides (as taught by McCloskey) using ligation techniques that were well-known in the art.

First, a POSA would have had a reason to consider McCloskey and its disclosure of dual tagging because Linnarsson and McCloskey are analogous art that focus on significantly the same subject matter. Both references describe methods of genetic analysis of single cells whereby polynucleotides are annealed to oligonucleotides comprising tag sequences (Linnarsson's CDS and TSO; McCloskey's encoding oligonucleotide) and primer extension takes place. Subsequently, PCR amplification is performed and the results are sequenced. EX1003 at 3:25-4:2; EX1015 at 2:27-3:3; EX1004 at Fig. 1. They are thus in the same field of endeavor and are based on the same approaches.

Second, a POSA would have been motivated to combine Linnarsson's use of a cell tag with McCloskey's molecule tag to distinguish individual molecules within

a sample because this approach addresses the amplification bias issue set forth in Linnarsson. EX1002 ¶¶ 171-174. Specifically, Linnarsson discloses that prior to sequencing, the "cDNA library can be further amplified, e.g. by PCR, to obtain a sufficient quantity of cDNA for sequencing." EX1003 at 21:21-22; EX1015 at 9:4-6; *see also* EX1002 ¶¶ 80-84 (explaining how PCR uses DNA polymerase and other reagents to synthesize new strands of DNA from template strands of DNA or RNA). But, as Linnarsson explains, "as the cDNA of each cell must be amplified to an amount that can be reasonably handled for the subsequent analysis, there is potential amplification bias." EX1003 at 2:3-13; EX1015 at 2:6-18. That is, certain molecules within the sample may be preferentially amplified relative to the other molecules in the sample, which could lead to distortions in the relative amounts of molecules as compared to what was originally present in the sample prior to amplification.

McCloskey provides the exact solution to this problem, which is to tag the polynucleotides ***before amplification*** with a tag that distinguishes different molecules within a sample from one another. As McCloskey explains, this "***solve[s]*** this source-uncertainty problem for DNA sequences generated by standard PCR." EX1004.001; *see also id.* at .005 ("An additional benefit of our method…is that counting sequences with nonredundant barcodes provides a quantitative measure of template diversity."). As mentioned, the risks of amplification bias are

"especially high when limited amounts of template DNA are used," such as for single cell genomics or gene expression analysis in single cells. EX1003 at 2:3-15, 11:6-10; EX1015 at 2:6-21, 3:14-20; EX1004 at Abstract. A POSA therefore would have been motivated to incorporate McCloskey's teachings into Linnarsson's methods to detect true sample complexity/template diversity. EX1002 ¶ 174. As Dr. Cooper explains, the tagging approach used in McCloskey for this purpose was well-known and had been used repeatedly in the art since at least 2004. *See* EX1002 ¶¶ 71-77, 87-89.

Finally, while McCloskey introduces both the cell tag and the molecule tag through a primer extension reaction, it would have been nothing more than an obvious and routine design choice to instead introduce the molecule tag via ligation. As of the earliest priority date of the '013 Patent, it was well-known and routine in the art to attach tag sequences to other sequences by ligation. *See* EX1002 ¶ 144. The '013 Patent itself acknowledges this, explaining that a "variety of template-driven ligation reactions" are available and citing nine different prior art references to this effect. *See* EX1001 at 6:31-40. Likewise, in the Detailed Description of the Invention, the '013 Patent affirms that the "practice of the present invention may employ, unless otherwise indicated, conventional techniques" and that such "conventional techniques" include "polymer array synthesis, hybridization, ***ligation***, and detection of hybridization using a label." *Id.* at 13:40-47.

While this alone establishes that the use of ligation would have been nothing but a routine and obvious design choice, Patent Owner further confirmed in its prior litigation against Becton-Dickinson that one would not require "***any*** new, non-conventional attachment techniques" "to attach 'diverse' (*i.e.* different) labels" by ligation. EX1017.011-012 (emphasis in original). Patent Owner stated with respect to the Fodor Patents, for instance, that "the specification admits that 'in one embodiment' of the invention 'the label-tag is attached by ligation' and that 'methods of ligation will be known to those of skill in the art.'" *Id*; *see also* EX1016.020 (admitting that "primer binding [was] known and conventional," citing references "describing known ligation techniques"). Patent Owner attacked the Fodor Patents, which specifically claim attaching tags to polynucleotides by ligation, as claiming nothing but "well-known and conventional" elements. EX1029 at Claim 17.

Given the undisputedly well-known and routine nature of ligation to attach label tags to polynucleotides, a POSA would have had a reasonable expectation of success in ligating a second sequence to Linnarsson's CDS or TSO. For example, it would have been obvious to do this by using a linker molecule. A 2003 article ("Hug") discloses ligating tag sequences to a polynucleotide by hybridizing a linker to the target polynucleotide, which is designed to hybridize to another polynucleotide used as a tag such that the tag is then ligated to the target

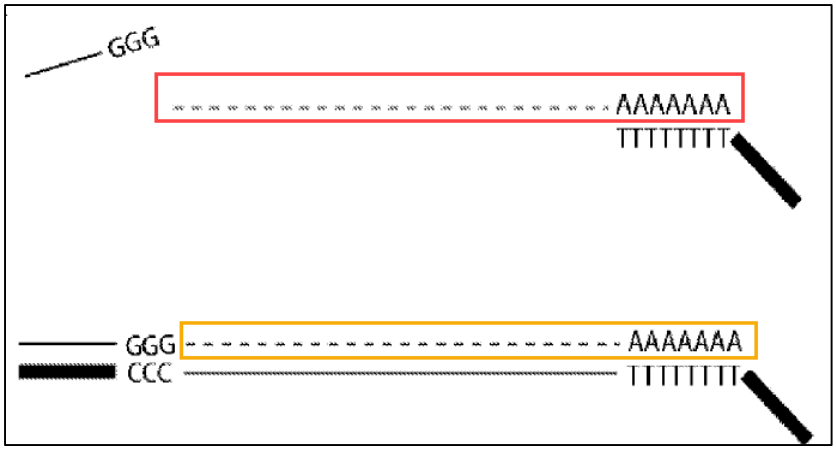
polynucleotide. EX1007.008; *see also* EX1002 ¶¶ 146-148 (explaining Hug's methods in detail).

A POSA could use Hug's method as one exemplary method to ligate the oligonucleotide comprising the tag sequences to Linnarsson's CDS or TSO. Specifically, a POSA would have designed a linker to hybridize to Linnarsson's CDS and/or TSO as well as the oligonucleotide, which would then be ligated to the CDS and/or TSO. *See* EX1002 ¶¶ 148-149. This approach was commonplace in the art at the time. *See id.* ¶¶ 148-150; *see also* EX1007.007; EX1024.005; EX1039.001-002; EX1040 at Fig. 2, ¶¶ 71-75, ¶¶ 235-239. As Dr. Cooper explains, this approach would be entirely equivalent to the method already disclosed by Linnarsson, except that instead of having the tag sequence be part of the CDS or TSO prior to extension, the second tag sequence is added to the CDS or TSO after extension. EX1002 ¶ 149. A POSA would have been motivated to utilize this design choice because it would allow for added flexibility in selecting tag sequences during an experiment. *See id.*

f. [1F] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises: a sample sequence corresponding to said sample polynucleotide of the plurality of sample polynucleotides;

Linnarsson discloses this claim element. As explained above in Sections IX.A.1.c-d, Linnarsson teaches generating a plurality of tagged polynucleotides (*i.e.*,

tagged double-stranded cDNA) from the plurality of sample polynucleotides (*i.e.*, sample mRNA). As shown in Figure 2, Linnarsson further discloses that the resulting double-stranded cDNA comprises one strand (boxed in orange) that is identical to the original sample mRNA (boxed in red):

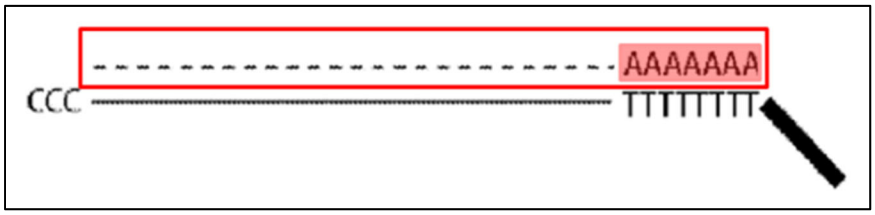


EX1003 at Fig. 2. Indeed, because the cDNA is synthesized from the sample mRNA using reverse transcription, one strand of each cDNA is necessary identical to the sample mRNA. *See* EX1002 ¶ 154.

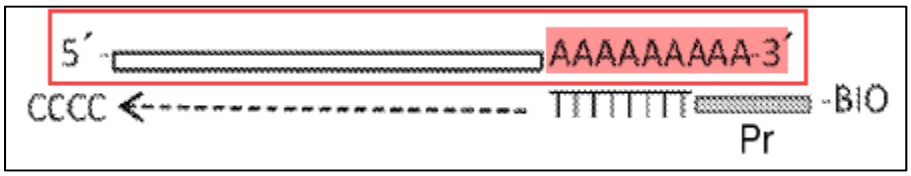
A POSA would have further concluded that a cDNA synthesized from a sample mRNA comprises the same sample sequence. EX1002 ¶ 154. While mRNA comprises uracil and DNA comprises thymine, any POSA in the field of gene analysis would have understood that uracil and thymine are functionally equivalent because they code for the same protein. *Id.*

Alternatively, Linnarsson discloses that the tagged cDNA comprises a sequence from the sample mRNA—a poly-A tail. Linnarsson discloses that the

sample mRNA comprises a poly-A tail. EX1003 at Fig. 2; EX1015 at Fig. 2; EX1002 ¶ 155. A "poly(A) tail" is a sequence of adenosine nucleotides found at the 3' end of mRNA molecules, such as the sample polynucleotides. *See* EX1002 ¶ 61. Figure 2 of Linnarsson, reproduced and annotated below, shows an mRNA molecule (boxed in red) with its poly-A tail (highlighted in red):



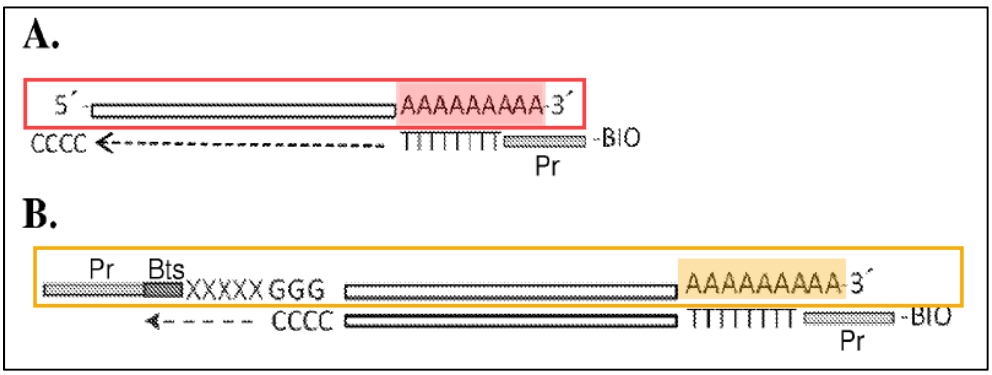
EX1003 at Fig. 2; EX1015 at Fig. 2; EX1003 at 4:14 (describing Fig. 2), 24:13-20 (same); EX1015 at 10:8, 13:8-18. Linnarsson's Figure 11 shows the same:



EX1003 at Fig. 11(A); EX1015 at Fig. 2; *see also* EX1002 ¶ 154.

Linnarsson discloses that after reverse transcription, the resulting double-stranded cDNA has a poly-A tail identical to the poly-A tail of the sample mRNA. Specifically, Linnarsson at Figure 2 discloses the resulting double-stranded cDNA, whereby one strand of the double-stranded cDNA (boxed in orange) shares the same sequence as the original mRNA strand (boxed in red), including a poly-A tail (highlighted in orange) that is the same as the poly-A tail (highlighted in red) of the original sample mRNA strand:

EX1003 at Fig. 2; EX1015 at Fig. 2. Figure 11 of Linnarsson discloses the same:



EX1003 at Fig. 11(A) and (B); EX1015 at Figs. 2, 3.

Therefore, Linnarsson discloses that each tagged polynucleotide (*i.e.*, tagged double-stranded cDNA and/or amplified copies) comprises a sample sequence (*i.e.*, the sample mRNA sequence and/or the poly-A tail) corresponding to said sample polynucleotide (*i.e.*, sample mRNA) of the plurality of sample polynucleotides.

g. [1G] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises:…a first tag sequence distinguishing said sample polynucleotide from sample polynucleotides from other cells; and

Linnarsson discloses this claim element. EX1002 ¶¶ 158-161. Specifically, Linnarsson discloses that the tag sequence comprises a first tag

sequence associated with the single cell from which the sample polynucleotide is derived, wherein that first tag sequence distinguishes the sample polynucleotide from sample polynucleotides from other cells. Linnarsson discloses "tagged reverse transcriptase is performed on each sample to produce cDNA," which allows for "identification of expressed genes and identification of ***cells from which they originated***." EX1003 at 4:8-13; EX1015 at 9:35-10:6. Linnarsson also explains that the cell-tags present in the CDS and/or the TSO and incorporated into the cDNA provide "a distinct tag, or a distinct combination of tags, such that…the tag can be used to identify ***which single cell from each cDNA sample originated***" and so that "each cDNA sample can be linked to a single cell…." EX1003 at 19:5-11; EX1015 at 7:26-34.

h. [1H] wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises:…a second tag sequence distinguishing said sample polynucleotide from other sample polynucleotides from said cell;

The combination of Linnarsson and McCloskey in view of the knowledge of the skilled artisan teaches this claim element for the reasons set forth above. *See supra* Section IX.A.1.e.

i. [1I] amplifying said tagged polynucleotide, thereby generating a plurality of amplified polynucleotides corresponding to the tagged polynucleotide; and

Linnarsson discloses this claim element. EX1002 ¶¶ 178-182. According to Linnarsson, after reverse transcription is performed on the mRNA molecules (*i.e.*, sample polynucleotides) to generate cDNA molecules (*i.e.*, tagged polynucleotides), the plurality of tagged cDNA are then amplified to generate copies of each tagged double-stranded cDNA. EX1003 at 10:20-22; EX1015 at 3:2-3; *see also* EX1003 at 4:11-12; EX1015 at 10:2-4. This amplification would generate a plurality of amplified polynucleotides corresponding to the tagged polynucleotides.

j. [1J] sequencing said plurality of amplified polynucleotides to determine sequences of the amplified polynucleotides corresponding to the sample sequence, the first tag sequence, and the second tag sequence of the tagged polynucleotide; and

Linnarsson discloses this claim element. EX1002 ¶¶ 183-187. Linnarsson teaches that after generating the tagged polynucleotides, "sequencing is performed" thereon. *See* EX1003 at 4:8-13, 19:28-32, 21:13-25 ("This cDNA library can be sequenced to provide an analysis of gene expression in single cells or in a plurality of single cells."); EX1015 at 8:22-28, 8:33-9:10, 9:35-10:6. As is standard, "sequencing" the amplified polynucleotides would determine the sequences of the polynucleotides corresponding to the sample sequence, the first tag sequence, and the second tag sequence of each tagged polynucleotide.

k. [1K] using the sequences determined in step (d) to count sample polynucleotides for multiple different sample polynucleotides of multiple different single cells of said plurality of cells.

The combination of Linnarsson and McCloskey discloses this claim element. As discussed above in Sections IX.A.1.d-e and g-h, the combination of Linnarsson and McCloskey discloses and renders obvious using a first tag to identify the single-cell origin of tagged polynucleotides, and using a second tag to uniquely identify each tagged polynucleotides.

Further, McCloskey discloses that these tags allow determining the precise count of sample polynucleotides in each sample single-cell. McCloskey teaches that by evaluating the determined sequences that "counting sequences with nonredundant barcodes provides a quantitative measure of template diversity." EX1004.006. Thus, as was routine, McCloskey's method provides that counting unique molecular tags would provide a count of sample polynucleotides. *See supra* Section VII.B. Further, an exemplary description of how the combination of Linnarsson and McCloskey teaches counting sample polynucleotides is in Dr. Cooper's accompanying declaration. EX1002 ¶¶ 240-242.

Indeed, as discussed in Section IX.A.1.a. and in the accompanying expert declaration of Dr. Cooper, the use of molecular tags for counting sample polynucleotides was well-known and routine not just as of the earliest priority date of the '013 Patent, but as of 2004. *See* EX1002 ¶ 72. The specification of the

’013 Patent provides that any “convenient method for tagging and/or counting individual nucleic acid molecules with unique tags may be employed.” EX1001 at 22:47-49; *see also* EX1002 ¶¶ 72, 74, 76 (describing prior art counting techniques cited by the ’013 Patent).

Thus, the combination of Linnarsson and McCloskey discloses and renders obvious using the first and second tag to count sample polynucleotides for multiple different sample polynucleotides of multiple different single cells of said plurality of cells.

2. Claim 2: The method of claim 1, wherein said plurality of sample polynucleotides comprise messenger ribonucleic acid (mRNA).

Disclosed. *See supra* Section IX.A.1.b.

3. Claim 3: The method of claim 2, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to said mRNA and (ii) extending said first oligonucleotide tag using said mRNA as a template to generate complementary DNA (cDNA).

Disclosed. *See supra* Section IX.A.1.d.

4. Claim 4: The method of claim 3, wherein step (B) comprises (i) ligating said second oligonucleotide tag to said cDNA.

For the same reason as explained above in Section IX.A.1.e, and because a POSA would know that the said “extension product” is “said cDNA,” this claim is rendered obvious by the combination of Linnarsson and McCloskey in view of the knowledge of a POSA. *See supra* Section IX.A.1.e; EX1002 ¶ 201.

5. Claim 5: The method of claim 3, wherein said hybridizing comprises hybridizing a poly-dT sequence of said first oligonucleotide tag to a poly-dA sequence of said mRNA.

Disclosed. *See supra* Section IX.A.1.d.

6. Claim 6: The method of claim 1, wherein said plurality of cells is provided in a plurality of wells.

As explained in Section IX.A.1.a, Linnarsson discloses that plurality of single cells are placed in separate wells. *See* EX1003 at 4:8-10; EX1015 at 9:35-10:3. Linnarsson further discloses that single cells "can be placed in any suitable reaction vessel in which single cells can be treated individually. For example a 96 well-plate, such that each single cell is placed in a single well." EX1003 at 13:20-22; EX1015 at 3:36-4:2; EX1002 ¶ 204.

7. Claim 7: The method of claim 1, wherein said first oligonucleotide tag comprises a sequence that is configured to hybridize to said sample polynucleotides.

Disclosed. *See supra* Section IX.A.1.d.

8. Claim 8: The method of claim 1, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to said sample polynucleotide and (ii) extending said first oligonucleotide tag or said sample polynucleotide or both to provide said extension product.

Disclosed. *See supra* Section IX.A.1.d.

9. Claim 9: The method of claim 1, wherein step (b) comprises randomly associating said plurality of sample polynucleotides with said plurality of oligonucleotide tags.

Linnarsson discloses a method for adding a plurality of oligonucleotide tags to a mixture containing sample polynucleotides to generate tagged polynucleotides. *See* EX1003 at 18:28-29 ("the CDS and/or the TSO can be added to each individual mRNA sample during cell lysis, as this will aid hybridization of the oligonucleotides."); EX1015 at 7:11-13. A POSA would have understood that by adding the oligonucleotide tags to a mixture containing sample polynucleotides under the conditions disclosed in Linnarsson, the tags would randomly associate with the sample polynucleotides to generate the plurality of tagged polynucleotides. EX1007.002 ("hybridization of DNA molecules with identical sequences is competitive and therefore, the tags are selected randomly"); EX1002 ¶ 209. Indeed, Linnarsson teaches that tags attach to mRNA nonspecifically; for example, every CDS attaches to mRNA by hybridizing with the poly-A tail, regardless of its unique tag sequence. *See supra* Section IX.A.1.c.

10. Claim 10: The method of claim 1, wherein said second tag sequences of said plurality of oligonucleotide tags are random sequences.

As discussed above in Section IX.A.1.h, the combination of Linnarsson and McCloskey teaches and renders obvious claim 10.

As discussed, McCloskey discloses a second tag sequence that is a "randomly generated barcode," which is used to distinguish between polynucleotides arising from the same single-cell. EX1004.003. McCloskey teaches that this second tag

"barcode is a random sequence of the four deoxyribonucleotides. The number of distinguishable barcodes in a population of oligonucleotides used in a reaction is determined by the number of **random bases, *n***." EX1004.004; EX1002 ¶ 211.

11. Claim 11: The method of claim 1, wherein step (c) comprises generating said plurality of amplified polynucleotides using polymerase chain reaction (PCR).

According to Linnarsson, after "synthesizing a first strand of cDNA from the mRNA in each individual mRNA sample" and "incorporating a tag," PCR amplification is performed to generate "a cDNA library comprising double-stranded cDNA"—*i.e.*, amplified polynucleotides. *See* EX1003 at 10:18-22; EX1015 at 2:33-3:3; *see also* EX1003 at 20:3-6; EX1015 at 8:7-9; EX1002 ¶ 214.

12. Claim 12: The method of claim 1, wherein substantially every sample polynucleotide of said plurality of sample polynucleotides is associated with the same first tag sequence.

As discussed above, Linnarsson discloses a cell-tag (*i.e.*, the "first tag sequence") that identifies the single-cell origin of sample polynucleotides. *See supra* Section IX.A.1.d.; EX1003 at 19:5-11; EX1015 at 7:28-34. It would have been obvious to tag substantially every sample polynucleotide with the same cell-tag.

First, a POSA would have been motivated to tag substantially every sample polynucleotide with the same cell-tag.[3] Linnarsson discloses using a cell-tag to identify the single-cell origin of sample polynucleotides. *See supra* Section IX.A.1.g. A POSA would have understood that tagging every sample polynucleotide (as opposed to only some of the sample polynucleotides) would allow one to maximize the amount of information gleaned from each cell. EX1002 ¶ 217.

Second, a POSA would have reasonably expected successfully tagging substantially every sample polynucleotide with the same cell-tag. The '013 Patent does not disclose any special methods or techniques for ensuring that substantially every sample polynucleotide becomes associated with the same first tag sequence. Just the opposite, the '013 Patent acknowledges it was known in the art to have "***each*** polynucleotide in the sample…tagged with a MID" by using a number of "MID tags…many times greater than the actual number of molecules to be analyzed." EX1001 at 22:59-23:2 (citing EX1035). The Brenner 2000 prior art article (EX1035), cited in the '013 patent specification, teaches, for instance, using over 100 times more tags than sample polynucleotides to accomplish this. *See* EX1001

[3] Claim 12 depends from claim 1, which recites "***a cell*** of the plurality of cells comprising ***a plurality of sample polynucleotides***." Therefore, the recited "every sample polynucleotide of ***said plurality of sample polynucleotides***" in claim 12 originates from "a cell" (*i.e.*, the same single-cell) as recited in claim 1.

at 22:66-23:2; EX1035.003-004. Consistent with this, as set forth in the accompanying declaration of Dr. Cooper, a POSA would have readily understood that increasing the amount of oligonucleotide tags relative to the number of sample molecules would drive the tagging reaction to completion so that every sample polynucleotide is associated with the same first tag sequence. *See* EX1002 ¶ 218. Achieving the appropriate concentrations of oligonucleotide tags for this purpose, Dr. Cooper explains, would have been nothing more than a straightforward optimization. *Id.*

13. Claim 19: The method of claim 1, wherein an oligonucleotide tag of said plurality of oligonucleotide tags comprises a sequencing adaptor.

Linnarsson discloses an oligonucleotide tag comprising a sequencing adapter. As discussed above, Linnarsson discloses two oligonucleotide tags, a TSO and a CDS. *See supra* Section IX.A.1.c. Linnarsson teaches that these tags comprise an amplification primer sequence ("APS"). EX1003 at 19:18-23; EX1015 at 8:9-15. This APS, which is incorporated into the oligonucleotide tag, may be designed to be a sequencing adapter. The "APS may be designed to facilitate downstream processing of the cDNA library. For example, if the cDNA library is to be analyzed by a particular sequencing method…the 5' APS can be designed to be identical to the primers used in those sequencing methods." EX1003 at 19:28-32; EX1015 at

8:22-28; EX1002 ¶¶ 223-225. Thus, Linnarsson teaches that the TSO and/or CDS (oligonucleotide tags) comprise a sequencing adapter.

14. Claim 20: The method of claim 19, wherein step (d) comprises hybridizing said amplified polynucleotides or derivatives thereof to a solid support via said sequencing adaptor or derivative thereof.

Linnarsson discloses sequencing via "any high-throughput screening methods," such as the routine and commercially available method of SOLiD sequencing. EX1003 at 21:16-18; EX1015 at 9:19-22. As Dr. Cooper explains, as of the 2009 timeframe, SOLiD sequencing was a standard DNA sequencing platform. EX1002 ¶ 228; EX1045.007. The SOLiD platform, along with other standard sequencing platforms that were commercially available in the 2009 timeframe, such as Roche 454, utilize as a standard step in the sequencing protocol the immobilization of sample nucleotides to a solid support, which was a bead. EX1002 ¶¶ 227-228. In the SOLiD platform, this is achieved via a biotin-streptavidin interaction whereas in the Roche 454 platform this is achieved via hybridization. *See* EX1045.003; EX1002 ¶ 228.

Linnarsson further teaches that the APS, which is the sequencing adapter in the CDS or TSO tag, "can be identical to the SOLiD P1 primer, and/or a SOLiD P2 sequence inserted in the CDS, so that the P1 and P2 sequences required for SOLiD sequencing are integral to the amplified library." EX1003 at 19:31-20:2; EX1015

at 8:28-31; *supra* Section IX.A.13. Thus, Linnarsson contemplates that the tagged DNA should include sequences to facilitate DNA sequencing.

While Linnarsson used the SOLiD platform for sequencing, it further teaches that "any high-throughput screening method" can be used. EX1003 at 21:16-18; EX1015 8:35-37. As Dr. Cooper explains, this would have encompassed the prior art Roche 454 platform, which was widely used in the 2009 time frame. Because Linnarsson teaches that "any high-throughput screening method" can be used and because the Roche 454 platform was so well-known at the time, a POSA would have been motivated to use Roche 454 as a routine alternative to the SOLiD platform used in Linnarsson. Importantly, in the Roche 454 system, this entails hybridizing the DNA to a solid support bead via the sequencing adaptor. As one article that summarizes the Roche 454 technology explains, "the library fragments are mixed with a population of agarose beads whose surfaces carry oligonucleotides complementary to the 454-specific adapter sequences on the fragment library, so each bead is associated with a single fragment." EX1045.003; *see also id.* at 1045.004 at Fig. 1b ("Anneal sstDNA to an excess of DNA capture beads.").

15. Claim 21: The method of claim 20, wherein said solid support is a bead.

For the same reason as explained above with respect to claim 20, this claim is rendered obvious by the combination of Linnarsson, McCloskey, and the knowledge

of a POSA, as exemplified by Roche 454 and/or SOLiD sequencing. *See supra* Section IX.A.14; EX1002 ¶ 223.

16. Claim 22:

a. [22A] The method of claim 1, wherein step (e) comprises (i) using second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide and/or

As explained above in Sections IX.A.1.e. and IX.A.1.h., the combination of Linnarsson and McCloskey teaches a second tag, which is a unique molecule-tag sequence attached to each tagged polynucleotide (i.e., tagged cDNA) originating from the same single-cell, and then using the molecule-tag to distinguish one tagged polynucleotide from other sample polynucleotides originating from the same single-cell. Thus, by using the second tag sequence, one may determine that a plurality of amplified polynucleotides comprising the same molecule-tag are amplified from the same tagged polynucleotide.

b. [22B] The method of claim 1, wherein step (e) comprises…(ii) using sample sequences, first tag sequences, and second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide.

As explained above in Section IX.A.1.d.-h. and IX.A.1.k, the combination of Linnarsson and McCloskey teaches a dual tagging method, where the first tag is a cell tag and distinguishes between tagged sequences arising from different cells,

and the second tag is a molecule tag that distinguishes between samples arising from the same cell. Thus, by identifying a plurality of amplified cDNA with the same sample sequence, first tag sequence, and the second tag sequence, one would determine that that said plurality of amplified polynucleotides are amplified from the same said tagged polynucleotide.

17. Claim 23: The method of claim 1, wherein step (e) comprises (i) determining the number of different second tags sequences associated with said sample sequence, thereby estimating the number of sample polynucleotides having said sample sequence from said single cell and/or (ii) using second tag sequences of said plurality of amplified polynucleotides to provide a digital count of said sample polynucleotides.

As explained above in Sections IX.A.1.d.-h. and IX.A.1.k., the combination of Linnarsson and McCloskey teaches unique molecule-tag sequences attached to each tagged polynucleotide (i.e., tagged cDNA) originating from the same single-cell, and using the molecule-tag to distinguish one tagged polynucleotides from other sample polynucleotides originating from the same single-cell. Further, the molecule-tag can distinguish among tagged polynucleotides even if they contain the same sample sequence (i.e., sample mRNA sequence), and therefore are synthesized from identical sample mRNA. Thus, by counting the number of different molecule-tags attached to tagged cDNA having the same sequence, one can determine the number of identical sample mRNA molecules in the sample single-cell.

As explained by Dr. Cooper, such technique of determining the number of sample polynucleotide having the same sequence was routine and well-known in the art as of at least 2004. EX1002 ¶¶ 239-240; EX1007.001 (providing two exemplary methods "to calculate the number of molecules of a single mRNA species in a complex mRNA preparation.").

Thus, by using a molecule tag ("second tag sequences"), one may determine the number of sample polynucleotides, which comprise the same sequence, and originate from the same single-cell. Therefore, the combination of Linnarsson and McCloskey in view of the knowledge of a POSA renders obvious this claim.

18. Claim 24: The method of claim 1, wherein said plurality of sample polynucleotides are pooled with sample polynucleotides from other single cells of said plurality of cells prior to the generating of step (b).

As described above in Sections IX.A.1.a., IX.A.1.d., and IX.A.1.I., Linnarsson discloses a method whereby the plurality of tagged polynucleotides are pooled with tagged polynucleotides from other single cells. It would be obvious to a POSA that such pooling could occur prior to the generating of step (b).

For example, it would be a simple design choice in view of McCloskey to utilize the first tag to identify groups of cells, rather than single-cells. EX1002 ¶ 244. McCloskey broadly teaches that its "batch-stamp" can be used for a variety of purposes beyond simply tracking single cells. Specifically, McCloskey contemplates that in addition to single-cells, the first tag ("batch-stamp") may be

used generally to identify the "DNA source," such as "the patient or sample." EX1004.002. Thus, polynucleotides from numerous cells of the same ***patient*** may be pooled together prior to being tagged. Therefore, said plurality of sample polynucleotides would be pooled with sample polynucleotides from other single cells of said plurality of cells (from the same patient) prior to the generating of step (b) wherein the sample polynucleotides are tagged with a patient specific first tag and a molecule specific second tag.

19. Claim 25: The method of claim 1, wherein said plurality of tagged polynucleotides are pooled with tagged polynucleotides from other single cells of said plurality of cells prior to the amplifying of step (c).

As described above in Sections IX.A.1.a., IX.A.1.d., and IX.A.1.I., Linnarsson discloses a method whereby the plurality of tagged polynucleotides are pooled with tagged polynucleotides from other single cells prior to amplification. For example, Linnarsson provides a method including a step of "pooling the tagged cDNA samples and amplifying the pooled cDNA samples." EX1003 at 10:20-22; EX1015 at 3:1-2.

20. Claim 26: The method of claim 1, wherein said plurality of amplified polynucleotides are pooled with amplified polynucleotides from other single cells of said plurality of cells prior to the sequencing of step (d).

As described above in Section IX.A.19., Linnarsson discloses a method of pooling the plurality of tagged polynucleotides prior to amplification, which would

also be prior to sequencing. Further, it would have been obvious to a POSA that to pool amplified polynucleotides prior to the sequencing of step (d) because, as described, samples have been associated with unique cell and molecule tags, which allow one to identify each samples origin after pooling samples for processing (either amplification or sequencing). EX1002 ¶ 249. Thus, a POSA would have been motivated to pool samples because doing so increases efficiency and speed and reduces cost. *Id*. Indeed, the '013 Patent cites a 2009 prior art publication disclosing that pooling of tagged polynucleotides prior to sequencing was available as a commercial product offering for use with, for instance, Roche 454 sequencing. EX1063.002 ("different libraries are prepared with different MIDs, they can be amplified by emPCR and sequenced together…that the reads from each of the pooled libraries are identified by their MID tag.").

21. Claim 27: The method of claim 1, wherein step (e) comprises using first tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the single cell from which the sample sequences are derived based on amplified polynucleotides from the same cell having the same first tag sequence.

As described above in Section IX.A.1.f.-g. and IX.A.16., the combination of Linnarsson and McCloskey teaches and renders obvious a method of tagging whereby a first tag is a cell-tag that is unique to each single-cell. Thus, each amplified polynucleotide which contains the same first tag sequence must have

arisen from the same single-cell of origin. Therefore, the combination of Linnarsson and McCloskey teaches a method of using the first tag sequence of a plurality of amplified polynucleotides to correlate the sample sequence of said amplified polynucleotides with the single cell from which the sample sequences were derived based on all amplified polynucleotides from the same cell having the same first tag sequence. EX1002 ¶ 251.

22. Claim 28: The method of claim 1, wherein step (e) comprises using second tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the sample polynucleotide from which the sample sequences are derived based on amplified polynucleotides from the same sample polynucleotide having the same second tag sequence.

As described above in Section IX.A.1.f.-h., IX.A.1.k., and IX.A.16., the combination of Linnarsson and McCloskey teaches and renders obvious a method of dual tagging whereby the second tag is a molecule-tag that is unique to each sample polynucleotide, which allows for distinguishing between sample polynucleotides which arise from the same cell. Therefore, the combination of Linnarsson and McCloskey teaches and renders obvious a method of using second tag sequences of a plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the sample polynucleotide from which they were derived based on the amplified

polynucleotides from the same sample polynucleotide having the same second tag sequence. EX1002 ¶ 253.

B. Ground 2

As explained below, Claims 13-18 of the ’013 Patent would have been obvious to a POSA over the combination of Linnarsson, McCloskey, McCloskey II, and further in view the knowledge of a POSA.

1. Claim 13: The method of claim 1, wherein at least 90 percent of said plurality of tagged polynucleotides have a second tag sequence that is different from second tag sequences of the other tagged polynucleotides.

The combination of Linnarsson, McCloskey, and McCloskey II renders this claim obvious.

First, a POSA would have been motivated to tag each sample polynucleotide with a unique second tag sequence. As explained above, the combination of Linnarsson and McCloskey discloses a second tag sequence used for counting and distinguishing sample polynucleotides originating from the same single-cell. *See supra* Sections IX.A.1.f, h. A POSA would have understood that increasing the number of polynucleotides tagged with a unique second tag sequence increases the accuracy of counting, and would have thus been motivated to ensure that each polynucleotides is tagged with a unique second tag sequence. EX1002 ¶ 255.

Second, a POSA would have had a reasonable expectation of successfully tagging each polynucleotide with a unique second tag sequence. As explained

above in Section IX.A.12, it would have been obvious to ensure that each sample polynucleotide of said plurality of sample polynucleotide is tagged by using a large number of tags (*e.g.*, at least 100 times more) relative to the number of sample polynucleotides. *See also* EX1001 at 22:58-66 (admitting that it was known to ensure that "each polynucleotide in the sample…tagged with a MID…different from every other MID on every other polynucleotide in the sample" by using a "number of distinct MID tags…many times greater than the actual number of molecules to be analyzed.") (citing EX1035).

Furthermore, it would have been obvious to design molecular tags to have a sufficient number of randomly selected nucleotides to ensure that each sample polynucleotide is tagged with a ***unique*** second tag sequence. McCloskey teaches that using second tag sequences each containing *n* number of random base pairs "enables one to distinguish 4^n allele copies per reaction." EX1004.004. It would have been a simple design choice to design the second tag sequences with appropriate number of nucleotides to ensure sufficient number of distinct second tag sequences in the reaction mixture (e.g., at least 100 times for than the number of sample polynucleotides) such that each sample polynucleotide is uniquely tagged. EX1002 ¶ 256.

For example, McCloskey II teaches using molecular-tags each comprising 30 random base pairs, which would allow the creation of 4^{30} (1.15×10^{18}) unique second

tag sequences. EX1055 at [0021] ("the length of the second sequence is between 3 and 30 nucleotides"). Linnarsson also teaches that its tag sequence can comprise at least 20 nucleotides, which would allow the creation of 4^{20} (1.10×10^{12}) unique barcodes. Using molecular tags comprising 30 or 20 nucleotides would more than ensure that each sample polynucleotide from a single cell is tagged with a unique tag sequence. A typical mammalian cell comprises approximately between 50,000 to 360,000 mRNA molecules. *See* EX1002 at ¶ 257.

Thus, the combination of Linnarsson, McCloskey, McCloskey II, and the knowledge of a POSA renders obvious claim 13.

2. Claim 14: The method of claim 1, wherein substantially every tagged polynucleotide of said plurality of tagged polynucleotides has a second tag sequence that is different from second tag sequences of the other tagged polynucleotides.

For the same reason as explained above with respect to claim 13, this claim is rendered obvious by the combination of Linnarsson and McCloskey II. *See supra* Section IX.B.1.

3. Claim 15: The method of claim 1, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is larger than the number of sample polynucleotides.

The combination of Linnarsson and McCloskey II discloses and renders obvious claim 15. As discussed above in Section IX.B.1., a POSA would have found it obvious to use more unique second tag sequences than sample

polynucleotides so that each sample polynucleotide tagged with a unique second tag sequence.

4. Claim 16: The method of claim 15, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is at least ten times the number of sample polynucleotides.

The combination of Linnarsson and McCloskey II discloses and renders obvious claim 16. As discussed above in Section IX.B.1., a POSA would have found it obvious to use at least ten times more unique second tag sequences than sample polynucleotides so that each sample polynucleotide tagged with a unique second tag sequence.

5. Claim 17: The method of claim 16, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is at least one hundred times the number of sample polynucleotides.

The combination of Linnarsson and McCloskey II discloses and renders obvious claim 17. As discussed above in Section IX.B.1., a POSA would have found it obvious to use at least 100 times more unique second tag sequences than sample polynucleotides so that each sample polynucleotide tagged with a unique second tag sequence.

6. Claim 18: The method of claim 1, wherein said plurality of oligonucleotide tags comprises at least 200,000 different second tag sequences.

The combination of Linnarsson and McCloskey II renders obvious this claim. As discussed above in Section IX.B.1., a POSA would have found it obvious to generate a massive number of second tags such that there are many times more second tags than sample polynucleotides. For example, McCloskey II teaches a method where a POSA easily design second tag sequences comprising 30 random base pairs, which would allow the creation of 1.15×10^{18} different second tag sequences. Moreover, a POSA would have been motivated to use more than 200,000 unique second tag sequences. As discussed above, a typical mammalian cell comprises several hundred thousand mRNA molecules. *Supra* Section IX.B.1; EX1002 ¶ 264. Thus, because it would have been obvious to use 100 times more second tags, it would have been obvious to use many times more than 200,000 different second tags.

Thus, the combination of Linnarsson and McCloskey renders obvious this claim.

X. THE BOARD SHOULD REACH THE MERITS OF THIS PETITION

A. Section 325(d) Does Not Apply Here, and Even if it Does, the Examiner's Material Errors Warrant Reaching the Merits of This Petition

In deciding whether to exercise its discretion to deny institution under 35 U.S.C. § 325(d), the Board applies a two-part framework that considers six non-exclusive factors. *See Becton, Dickinson & Co. v. B. Braun Melsungen AG*,

IPR2017-01586, Paper 8 at 17-18 (P.T.A.B. Dec. 15, 2017) (precedential). As documented below, the same art and arguments presented herein were not previously presented to the Office in any meaningful or relevant way. In addition, the Examiner erred by not considering the relevant teachings in McCloskey alone or in combination with Linnarsson. As such, application of *Becton* overwhelmingly confirms that the Board should not exercise its discretion to deny institution under § 325(d).

1. Factors (a), (b) and (d): The Office Has Not Considered the Same or Substantially the Same Art or Arguments

McCloskey was not in front of the Examiner during prosecution of the '013 patent. Although the Applicant identified McCloskey II and Linnarsson in an IDS during the prosecution, the Examiner did not cite or discuss McCloskey II or Linnarsson in any rejection or in any remarks in response to a rejection. EX1005.005, 142, 112-117, 130-132.

Accordingly, with respect to McCloskey, McCloskey II, and Linnarsson, "the overlap between the arguments made during examination" and the arguments made in this petition is minimal, as ***no arguments*** were made during examination with respect to any of these references. *See 3Shape A/S v. Align Tech., Inc.*, IPR2019-00160, Paper 9 at 39-42 (P.T.A.B. June 11, 2019) (declining to apply § 325(d) where there was no evidence that the examiner substantively considered references that were of record during prosecution); *Microsoft Corp. v. Parallel Networks Licensing,*

LLC, IPR2015-00486, Paper 10 at 15 (P.T.A.B. July 15, 2015) (declining to exercise § 325(d) discretion because prior art reference at issue was only listed on IDS and "there is no evidence that the Examiner considered the particular disclosures cited by [Petitioner] in the Petition").

Linnarsson and McCloskey II are cited on the face of the '013 Patent. Mention of references in the specification, however, does not justify denial under § 325(d), particularly where, as here, neither were cited nor discussed by the Examiner during prosecution. *See Limelight Networks, Inc. v. Akamai Techs., Inc.*, IPR2016-01631, 2016 WL 8115506, at *5 (P.T.A.B. Dec. 30, 2016) (declining to exercise § 325(d) discretion because the Board was "not persuaded that merely referencing [references] in the '102 patent's specification necessarily is the same as presenting arguments with respect to those references"); *Weber, Inc. v. Provisur Techs., Inc.*, IPR2019-01467, Paper 7 at 9-10 (P.T.A.B. Feb. 14, 2020); *3Shape A/S and supra Microsoft*.

2. Factors (c), (e), and (f): The Examiner Erred by Not Considering the Art and Relevant Teachings Herein

Even if the Board determines that the same or substantially the same art or arguments were before the Office, the Examiner materially erred by not considering the relevant teachings in Linnarsson, McCloskey, and McCloskey II.

In the Notice of Allowance, the Examiner did not discuss or cite any passage or disclosure of Linnarsson or McCloskey II. EX1005.130-132. In fact, as

documented above, the Examiner committed major error in Examination by erroneously stating that the prior art fails to teach a dual tagging approach when, in fact, this approach is plainly taught in both McCloskey and McCloskey II. *See supra* Part V.D.

In such a case, where "the record of the Office's previous consideration of the art is not well developed...a petitioner may show the Office erred by overlooking something persuasive under factors (e) and (f)." *Advanced Bionics, LLC v. MED-EL Elektromedizinische Gerate GmbH*, IPR2019-01469, Paper 6 at 10 (P.T.A.B. Feb. 13, 2020); *see Kayak Software Corp. v. Int'l Bus. Machs. Corp.*, CBM2016-00075, Paper 16 at 11 (P.T.A.B. Dec. 15, 2016) (informative) ("where the prior art at issue was only cursorily considered that can weigh against exercising the discretion"). Moreover, the fact that neither Linnarsson nor McCloskey II was "the basis of a rejection weighs strongly against exercising [the Board's] discretion to deny institution." *Cellco P'ship v. Huawei Device Co., Ltd.*, IPR2020-01117, Paper 10 at 13 (P.T.A.B. Feb. 3, 2021).

Here, the Examiner erred by failing to acknowledge any teachings in Linnarsson or McCloskey II. *Amber.IO, Inc. v. 72LUX, Inc.*, IPR2020-00015, Paper 8 at 20 (P.T.A.B. Apr. 1, 2020) (finding that the Examiner materially erred by failing to consider prior art references listed on IDS, when the Petitioner demonstrated that the references teach significant elements of the challenged

claims). The Board should reach the merits of this petition and consider these relevant teachings in the first instance.

B. The *Fintiv* Factors Favor Institution

The factors set forth in *Apple Inc. v. Fintiv, Inc.*, IPR2020-00019, Paper 11 (P.T.A.B. Mar. 20, 2020) ("*Fintiv*"), for weighing discretionary denial when there is parallel, co-pending district court litigation, also favor institution.

The **first *Fintiv* factor**—whether the court granted a stay or evidence exists that one may be granted if a proceeding is instituted—is neutral. If instituted, Petitioner will seek stay in the district court litigation. The court's decision will come after institution and is "based on a variety of circumstances and facts beyond [the Board's] control and to which the Board is not privy." *Sand Revolution II, LLC v. Continental Intermodal Grp. – Trucking, LLC*, IPR2019-01393, Paper 24 at 7 (P.T.A.B. June 16, 2020) (informative); *see Western Digital Corp. v. Kuster*, IPR2020-01410, Paper 13 at 9-10 (P.T.A.B. Feb. 17, 2021). Moreover, the District of Delaware grants the majority of motions to stay pending IPR. EX1023; *see also, e.g.*, *International Business Machines Corp. v. Rakuten Inc.*, No. 21-461-GBW, 2022 WL 17848779 (D. Del. Dec. 22, 2022) (granting defendant's motion to stay after the Board instituted IPRs for the asserted patents). Thus, this factor is at least neutral.

The **second *Fintiv* factor**—proximity of the court's trial date to the Board's projected statutory deadline for a final written decision—favors institution. Currently, the trial in the district court litigation is scheduled to start on December 2, 2024, which will be after the anticipated final written decision in this IPR. EX1018.

But more realistically, it is far from certain that the district court trial will proceed as planned. There is a reasonable chance that the litigation may be stayed upon institution and/or ordinary delays in district court litigation may occur over the next 18 months. Indeed, the median time to trial for civil cases in the District of Delaware is 33.7 months, which would place trial in July 2025. EX1019.014. Thus, this factor favors institution.

The **third *Fintiv* factor**—investment in the parallel proceeding by the court and the parties—favors institution. The Board has found that the third factor favors institution where in the parallel litigation, "the District Court has not issued any substantive orders," "no claim construction orders have issued," and "discovery is not complete, because … neither party had taken any depositions." *Snap, Inc. v. SRK Tech. LLC*, IPR2020-00820, Paper 15 at 10–11 (P.T.A.B. Oct. 21, 2020). This is all true for the district court litigation.

Fact discovery is ongoing. Further, expert discovery or dispositive motion briefing will not be completed until well after the institution deadline. EX1018.

No claim construction brief has been filed by either party, and the claim construction hearing will not occur until February 21, 2024. EX1018. Furthermore, "the most burdensome parts of the case—filing and responding to pretrial motions, preparing for trial, going through the trial process, and engaging in post-trial motions practice—all lie in the future." *CyWee Grp. Ltd. v. Samsung Elecs. Co. Ltd.*, No. 2:17-cv-00140-WCB-RSP, 2019 WL 11023976 (E.D. Tex. Feb. 14, 2019). Thus, this third factor favors institution.

The **fourth *Fintiv* factor**—overlap between issues raised in the petition and in the parallel proceeding—favors institution. If instituted, Petitioner will stipulate to not pursuing, in the district court litigation, invalidity arguments based on any instituted IPR ground.

The **fifth *Fintiv* factor**—whether the Petitioner and the defendant in the parallel proceeding are the same party—is neutral. Petitioner is the defendant in the district court litigation, but that is often the case for a patent asserted in district court and challenged at the Board.

Finally, the **sixth *Fintiv* factor**—other circumstances that impact the Board's exercise of discretion, including the merits—also favors institution. This Petition presents a compelling merits case that all claims of the '013 Patent are unpatentable, and therefore this factor weighs in favor of institution. *Supra* Section VIII; *Fintiv*, IPR2020-00019, Paper 11 at 14–15.

Thus, in view of the facts presented in this Petition, the *Fintiv* factors do not support discretionary denial.

XI. CONCLUSION

Petitioner respectfully requests cancellation of the Challenged Claims.

Dated: May 25, 2023

Respectfully submitted,

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APPENDIX: CHALLENGED CLAIM LISTING

No.	Limitation
1	A method for multiplexed analysis of nucleic acids from single cells, the method comprising: (a) providing a sample comprising a plurality of cells, wherein a single cell of the plurality of cells comprises a plurality of sample polynucleotides; (b) performing combinatorial tagging to generate a plurality of tagged polynucleotides from said plurality of sample polynucleotides and a plurality of oligonucleotide tags, wherein a tagged polynucleotide of the plurality of tagged polynucleotides is generated by: (A) providing an extension product by primer extension using a first oligonucleotide tag and a sample polynucleotide of said plurality of sample polynucleotides, and (B) ligating a second oligonucleotide tag to said extension product, and wherein said tagged polynucleotide of the plurality of tagged polynucleotides comprises: (i) a sample sequence corresponding to said sample polynucleotide of the plurality of sample polynucleotides; (ii) a first tag sequence distinguishing said sample polynucleotide from sample polynucleotides from other cells; and (iii) a second tag sequence distinguishing said sample polynucleotide from other sample polynucleotides from said cell; (c) amplifying said tagged polynucleotide, thereby generating a plurality of amplified polynucleotides corresponding to the tagged polynucleotide; and (d) sequencing said plurality of amplified polynucleotides to determine sequences of the amplified polynucleotides corresponding to the sample sequence, the first tag sequence, and the second tag sequence of the tagged polynucleotide; and

	(e) using the sequences determined in step (d) to count sample polynucleotides for multiple different sample polynucleotides of multiple different single cells of said plurality of cells.
2	The method of claim 1, wherein said plurality of sample polynucleotides comprise messenger ribonucleic acid (mRNA).
3	The method of claim 2, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to said mRNA and (ii) extending said first oligonucleotide tag using said mRNA as a template to generate complementary DNA (cDNA).
4	The method of claim 3, wherein step (B) comprises (i) ligating said second oligonucleotide tag to said cDNA.
5	The method of claim 3, wherein said hybridizing comprises hybridizing a poly-dT sequence of said first oligonucleotide tag to a poly-dA sequence of said mRNA.
6	The method of claim 1, wherein said plurality of cells is provided in a plurality of wells.
7	The method of claim 1, wherein said first oligonucleotide tag comprises a sequence that is configured to hybridize to said sample polynucleotides
8	The method of claim 1, wherein step (A) comprises (i) hybridizing said first oligonucleotide tag to said sample polynucleotide and (ii) extending said first oligonucleotide tag or said sample polynucleotide or both to provide said extension product.
9	The method of claim 1, wherein step (b) comprises randomly associating said plurality of sample polynucleotides with said plurality of oligonucleotide tags.
10	The method of claim 1, wherein said second tag sequences of said plurality of oligonucleotide tags are random sequences.
11	The method of claim 1, wherein step (c) comprises generating said plurality of amplified polynucleotides using polymerase chain reaction (PCR).
12	The method of claim 1, wherein substantially every sample polynucleotide of said plurality of sample polynucleotides is associated with the same first tag sequence.
13	The method of claim 1, wherein at least 90 percent of said plurality of tagged polynucleotides have a second tag sequence that is different from second tag sequences of the other tagged polynucleotides

14	The method of claim 1, wherein substantially every tagged polynucleotide of said plurality of tagged polynucleotides has a second tag sequence that is different from second tag sequences of the other tagged polynucleotides.
15	The method of claim 1, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is larger than the number of sample polynucleotides.
16	The method of claim 15, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is at least ten times the number of sample polynucleotides.
17	The method of claim 16, wherein said plurality of oligonucleotide tags comprises a number of different second tag sequences that is at least one hundred times the number of sample polynucleotides.
18	The method of claim 1, wherein said plurality of oligonucleotide tags comprises at least 200,000 different second tag sequences.
19	The method of claim 1, wherein an oligonucleotide tag of said plurality of oligonucleotide tags comprises a sequencing adaptor.
20	The method of claim 19, wherein step (d) comprises hybridizing said amplified polynucleotides or derivatives thereof to a solid support via said sequencing adaptor or derivative thereof.
21	The method of claim 20, wherein said solid support is a bead.
22	The method of claim 1, wherein step (e) comprises (i) using second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide and/or (ii) using sample sequences, first tag sequences, and second tag sequences of said plurality of amplified polynucleotides to determine that said plurality of amplified polynucleotides are amplified from said tagged polynucleotide.
23	The method of claim 1, wherein step (e) comprises (i) determining the number of different second tags sequences associated with said sample sequence, thereby estimating the number of sample polynucleotides having said sample sequence from said single cell and/or (ii) using second tag sequences of said plurality of amplified polynucleotides to provide a digital count of said sample polynucleotides.

24	The method of claim 1, wherein said plurality of sample polynucleotides are pooled with sample polynucleotides from other single cells of said plurality of cells prior to the generating of step (b).
25	The method of claim 1, wherein said plurality of tagged polynucleotides are pooled with tagged polynucleotides from other single cells of said plurality of cells prior to the amplifying of step (c).
26	The method of claim 1, wherein said plurality of amplified polynucleotides are pooled with amplified polynucleotides from other single cells of said plurality of cells prior to the sequencing of step (d).
27	The method of claim 1, wherein step (e) comprises using first tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the single cell from which the sample sequences are derived based on amplified polynucleotides from the same cell having the same first tag sequence
28	The method of claim 1, wherein step (e) comprises using second tag sequences of said plurality of amplified polynucleotides to correlate the sample sequences of said plurality of amplified polynucleotides with the sample polynucleotide from which the sample sequences are derived based on amplified polynucleotides from the same sample polynucleotide having the same second tag sequence.

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Respectfully submitted,

WEIL, GOTSHAL AND MANGES LLP

Dated: May 25, 2023

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CERTIFICATE OF SERVICE

I hereby certify that a true and correct copy of **PETITION FOR *INTER PARTES* REVIEW OF U.S. PATENT NO. 10,697,013 WITH PARSE BIOSCIENCES, INC'S EXHIBIT LIST, POWER OF ATTORNEY AND EXHIBITS 1001-1063** are being served on May 25, 2023, via Priority Express Mail (Fed Ex) pursuant to 37 C.F.R. § 42.105 and § 42.6(e) to the Patent Owner at the address below:

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