

UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE PATENT TRIAL AND APPEAL BOARD

PACIFIC BIOSCIENCES OF CALIFORNIA, INC.,
Petitioner

v.

THE CHINESE UNIVERSITY OF HONG KONG,
Patent Owner

PTAB Case No. IPR2024-00028
U.S. Patent No. 11,091,794

PATENT OWNER'S RESPONSE

TABLE OF CONTENTS

	Page
I. INTRODUCTION	1
II. THE '794 PATENT	4
A. Claim 1	5
B. Prosecution of Claim 1	7
C. Claim Construction.....	11
D. Person of Ordinary Skill in the Art	11
III. CLAIM 1 IS NOT OBVIOUS OVER GROUND 1.....	11
A. Flusberg, the '338 Patent and the PacBio-BAM do not Disclose or Render Obvious the Combination of Nucleotide Identity, Position, PW, IPD in an Input Data Structure of Claim 1.	12
1. Identity of the nucleotide	14
a. No reference includes “identity of nucleotide” with any other property recited in claim 1.....	17
b. The Petition’s other listed reason do not support unpatentability under Ground 1.....	21
c. “Sequence context” does not mean “identity of the nucleotide”	29
2. A POSA would not have included pulse width with the other properties of the claimed input data structure.	31
IV. OBJECTIVE INDICIA OF NONOBVIOUSNESS SHOW THAT THE INPUT DATA STRUCTURE OF CLAIM 1 WAS NOT OBVIOUS AT THE TIME OF THE INVENTION.	36
A. The Method of Claim 1 Solved the Long-Felt Unsolved Need for Direct Detection of DNA Modifications by SMRT Sequencing Without Bisulfite Conversion.....	37

TABLE OF CONTENTS (CONTINUED)

	Page
1. The need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing was a persistent need recognized by a POSA.	38
a. Bisulfite sequencing was the most common DNA methylation detection method and the recognized gold standard.	39
b. The drawbacks of bisulfite conversion/sequencing created a persistent long-felt need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing.	40
2. No one satisfied the long-felt need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing before the '794 patent.	44
3. The input data structure of claim 1 directly detects DNA methylation without bisulfite conversion through SMRT sequencing and produces comparable results.	45
B. PacBio Tried and Failed to Directly Detect DNA Methylation Without Bisulfite Conversion Through SMRT Sequencing	49
C. The Holistic Kinetic Model Was Praised by PacBio and others in the field.	50
D. PacBio Copied the Holistic Kinetic Model Disclosed in the Tse Paper.	54
a. PacBio had access to the invention through the Tse paper.	54
b. The PacBio product bears a striking similarity to the invention disclosed in the TSE paper.	55
c. After the TSE publication PacBio suddenly changed direction and adopted claim 1's input data structure.	59

TABLE OF CONTENTS (CONTINUED)

	Page
E. The Method of Claim 1 Had Unexpected Results Compared to Flusberg	62
V. PATENTABILITY OF CLAIMS 2-19	65
VI. CONCLUSION.....	66

EXHIBIT LIST

	Petitioner's Exhibits
No.	Description
1001	U.S. Patent No. 11,091,794 to Lo ("the '794 patent")
1002	Declaration of Dr. Christophe Dessimoz
1003	Curriculum Vitae of Christophe Dessimoz, Ph.D.
1004	U.S. Patent Application No. 16/995,607 File History
1005	U.S. Patent Application No. 17/379,544 File History
1006	Flusberg, et al., "Direct Detection of DNA Methylation During Single-molecule, Real-Time Sequencing," <i>Nature Methods</i> 7(6) (2010) ("Flusberg")
1007	U.S. Patent No. 9,175,338 to Flusberg et al. ("Flusberg-338")
1008	Clark, et al. "Direct Detection and Sequencing of Damaged DNA Bases" <i>Genome Integrity</i> 2 (2011) ("Clark")
1009	Loomis et al., "Sequencing the Unsequenceable: Expanded CGG-repeat Alleles of the Fragile X Gene," <i>Genome Research</i> 23(1) (2013) ("Loomis")
1010	Schadt, et al., "Modeling Kinetic Rate Variation in Third Generation DNA Sequencing Data to Detect Putative Modifications to DNA Bases," <i>Genome Research</i> 23(1) (2013) ("Schadt")
1011	BAM Format Specification for PacBio, PacBioFileFormats 5.0 Documentation, http://web.archive.org/web/20170916112547/http://pacbiofileformats.readthedocs.io/en/5.0/BAM.html (archived Sep. 16, 2017) ("PacBio-BAM")
1012	SAM/BAM Specification Working Group, Sequence Alignment/Map Format Specification, http://web.archive.org/web/20180613134937/http://samtools.github.io/hts-specs/SAMv1.pdf (archived Jun. 13, 2018) ("SAM/BAM Specification")

Petitioner's Exhibits	
No.	Description
1013	Davis et al., “Entering the Era of Bacterial Epigenomics with Single Molecule Real Time DNA Sequencing,” <i>Current Opinion in Microbiology</i> , 16(2) (2013) (“Davis”)
1014	<i>Take2 Tech, Ltd. v. Pac. Biosciences of Cal., Inc.</i> , DDE-1-22-cv-01595, Infringement Claim Chart for U.S. Patent No. 11,091,794 (“Take2 Infringement Contentions”)
1015	Feng, et al., “Detecting DNA modifications from SMRT Sequencing Data by Modeling Sequence Context Dependence of Polymerase Kinetic,” <i>PLoS Computational Biology</i> , 9(3) (2013) (“Feng”)
1016	“Detecting DNA Base Modifications Using Single Molecule, Real-Time Sequencing,” https://www.pacb.com/wp-content/uploads/2015/09/WP_Detecting_DNA_Base_Modifications_Using_SMRT_Sequencing.pdf (2012) (“PacBio White Paper”)
1017	Zou et al., “A Primer on Deep Learning in Genomics” <i>Nature Genetics</i> , 51(1) (Jan. 2019)
1018	U.S. Patent Pub. No. 2016/0326593 to Clement et al. (“Clement”)
1019	Pacific Biosciences Template Preparation and Sequencing Guide, https://www.pacb.com/wp-content/uploads/2015/09/Guide-Pacific-Biosciences-Template-Preparation-and-Sequencing.pdf (2015) (“PacBio Template Guide”)
1020	U.S. Patent Publication No. 2016/0208241 to Tsai et al. (“Tsai”)
1021	Pacific Biosciences kineticsTools Manual, http://web.archive.org/web/20170813085437/https://github.com/PacificBiosciences/kineticsTools/blob/master/doc/manual.rst (2018) (“kineticsTools manual”)
1022	Liu et al., “Detection of DNA Base Modifications by Deep Recurrent Neural Network on Oxford Nanopore Sequencing Data,” <i>Nature Comms.</i> 10(1) (Jun. 4, 2019) (“Q. Liu”)

Petitioner's Exhibits	
No.	Description
1023	Eid et al., "Real-time DNA sequencing from single polymerase molecules," <i>Science</i> 323(5910) (2009)
1024	Int. Appl. Publ. No. WO 2012/065043 ("Flusberg PCT")
1025	Jonas Korlach & Stephen W. Turner, "Single-Molecule Sequencing" in <i>Encyclopedia of Biophysics</i> (2013) ("Korlach")
1026	GenomeWeb, "PacBio Reveals Beta System Specs for RS; Says Commercial Release is on Track for First Half of 2011," https://www.genomeweb.com/sequencing/pacbios-q3-losses-widen-it-readies-launch-rs-system (2010) ("GenomeWeb")
1027	Jacob Schreiber et al., "Error Rates for Nanopore Discrimination among Cytosine, Methylcytosine, and Hydroxymethylcytosine along Individual DNA Strands," <i>Proceedings of the National Academy of Sciences</i> 110(47) (2013) ("Schreiber")
1028	Langlotz, A Roadmap for Foundational Research on Artificial Intelligence in Medical Imaging: From the 2018 NIH/RSNA/ACR/The Academy Workshop, <i>Radiology</i> 291(3) (Apr. 16, 2019) ("Langlotz")
1029	Miotto et al., "Deep Learning for Healthcare: Review, Opportunities and Challenges," <i>Briefings in Bioinformatics</i> , 19(6) (2018) ("Miotto")
1030	Tse et al., "Genome-wide Detection of Cytosine Methylation by Single Molecule Real-time Sequencing," <i>Proceedings of the National Academy of Sciences</i> , 118(5) (2021) ("Tse")
1031	Tiernan Ray, "AI is Changing the Entire Nature of Compute," <i>ZDNet</i> , http://web.archive.org/web/20190630155704/https://www.zdnet.com/article/ai-is-changing-the-entire-nature-of-compute/ (June 30, 2019) ("Ray")
1032	Wenger et al., "Accurate Circular Consensus Long-read Sequencing Improves Variant Detection and Assembly of a Human Genome," <i>Nature Biotechnology</i> , 37(10) (Aug. 12, 2019) ("Wenger")

Petitioner's Exhibits	
No.	Description
1033	Pacific Biosciences, PacBioFileFormats Documentation (Feb. 2022) ("PacBioFileFormats-2022")
1034	Lau et al., "LongISLND: <i>in silico</i> Sequencing of Lengthy and Noisy Datatypes," <i>Bioinformatics</i> , 32(24) (2016) ("Lau")
1035	Tahir et al., "iRNA-PseKNC(2methyl): Identify RNA 2'-O-methylation Sites by Convolution Neural Network and Chou's Pseudo Components," <i>Journal of Theoretical Biology</i> , 465 (2018) ("Tahir")
1036	DeepVariant Usage Guide, https://github.com/google/deepvariant/blob/42b9c777436d8306f6cb9d2e18da756fa370fcd8/docs/deepvariant-details.md ("DeepVariant")
1037	U.S. Patent Application No. 18/189,084 File History
1038	Liu et al., "Accurate Detection of m6A RNA Modifications in Native RNA Sequences," https://www.biorxiv.org/content/10.1101/525741v1 (Jan. 21, 2019) ("H. Liu")
1039	First Declaration of Nathaniel E. Frank-White
1040	Second Declaration of Nathaniel E. Frank-White
1041	Declaration of Mozhgan Novbakhtian
1042	GitHub, PacBio BAM 3.0, https://github.com/PacificBiosciences/PacBioFileFormats/blob/3.0/BAM.rst (Mar. 28, 2016)
1043	GitHub, PacBio BAM 5.0, https://github.com/PacificBiosciences/PacBioFileFormats/blob/5.0/BAM.rst (Mar. 7, 2017)
1044	PacBio Sequencing – How it Works

Patent Owner's Exhibits	
No.	Description
2001	Color Drawings of US 11,091,794 Downloaded from USPTO Patent Center
2002	Declaration of Dr. Peter Koo
2003	Dr. Peter Koo's CV
2004	Li et al. and 1000 Genome Project Data Processing Subgroup. 2009. "The Sequence Alignment/Map format and SAMtools," <i>Bioinformatics</i> 25(16): 2078-2079 ("Li")
2005	Suzuki et al., "AgIn: measuring the landscape of CpG methylation of individual repetitive elements," <i>Bioinformatics</i> , 32(19), 2016, 2911–2919 ("Suzuki")
2006	Zheng and Casari, "Feature Engineering for Machine Learning," First Edition, O'Reilly Media, Inc., Sebastopol, CA, 2018 (Excerpts)
2007	Web page (print friendly screen capture): Johns Hopkins University Center for Computational Biology, StringTie – Transcript assembly and quantification for RNA-Seq. URL: http://ccb.jhu.edu/software/stringtie/index.shtml . Web page visited January 23, 2024
2008	Web page (print friendly screen capture): Broad Institute Genome Analysis Toolkit. URL: http://gatk.broadinstitute.org/hc/en-us . Web page visited January 23, 2024
2009	Web page (print friendly screen capture): Broad Institute Genome Analysis Toolkit - HaplotypeCaller. March 4, 2020. URL: http://gatk.broadinstitute.org/hc/en-us/articles/360040096812-HaplotypeCaller . Web page visited January 23, 2024
2010	Web page: University of California at Santa Cruz, Genome Browser BAM Track Format. URL: https://genome.ucsc.edu/goldenPath/help/bam.html . Web page visited January 23, 2024

Patent Owner's Exhibits	
No.	Description
2011	Web page (print friendly screen capture): Git Hub HPC Training – Peak calling with MACS2. June 12, 2017. URL: https://hbctraining.github.io/Intro-to-ChIPseq/lessons/05_peak_calling_mac.html . Web page visited January 23, 2024
2012	YouTube Video: PacBio Data Deep Dive: A Closer Look at HiFi Sequencing. PacBio May 24, 2021. URL: https://www.youtube.com/watch?v=1aqnLQ-nwYk . Downloaded January 22, 2024. (Also available at https://www.pacb.com/videos/pacbio-data-deep-dive-a-closer-look-at-hifi-sequencing/)
2013	Screen Capture of Exhibit 2012 (YouTube) https://www.youtube.com/watch?v=1aqnLQ-nwYk
2014	Screen Capture of Exhibit 2012 (PacBio.com) https://www.pacb.com/videos/pacbio-data-deep-dive-a-closer-look-at-hifi-sequencing/
2015	Transcript of 9:50 – 11:45 of Exhibit 2012
2016	<i>Take2 Technologies Limited v. Pacific Biosciences of California, Inc.</i> , U.S. District Court for the District of Delaware, Case No. 5:23-cv-04166-EJD, Dkt. 8, Complaint (Public Redacted Version), December 14, 2022
2017	<i>Take2 Technologies Limited v. Pacific Biosciences of California, Inc.</i> , 5:23-cv-04166-EJD, Dkt. 8-2, Exhibit C to the Complaint – Transcript of PacBio Q4 2020 Earnings call on February 10, 2021 (available at https://www.fool.com/earnings/call-transcripts/2021/02/10/pacific-biosciences-of-californiapacb-q4-2020-ear/) (Complaint Ex. C)

	Patent Owner's Exhibits
No.	Description
2018	<i>Take2 Technologies Limited v. Pacific Biosciences of California, Inc.</i> , 5:23-cv-04166-EJD, Dkt. 8-2, Exhibit E to the Complaint –PacBio Children's Mercy Kansas City Press Release dated April 21, 2022 (available at https://www.pacb.com/press_releases/pacb-and-childrens-mercy-kansas-city-expand-collaboration-taking-a-multi-omics-approach-to-characterize-rare-disease/) (Complaint Ex. E)
2019	<i>Take2 Technologies Limited v. Pacific Biosciences of California, Inc.</i> , 5:23-cv-04166-EJD, Dkt. 8-2, Exhibit F to the Complaint - Screenshot PacBio Products - Epigenetics (available at https://www.pacb.com/products-and-services/applications/epigenetics/) (Complaint Ex. F)
2020	YouTube Video: "HiFi Sequencing: See What You've Been Missing." PacBio, November 2, 2021. URL: https://www.youtube.com/watch?v=d2an7LXhPSU . Downloaded January 19, 2024. (Also available at https://www.pacb.com/videos/hifi-sequencing-see-what-youve-been-missing/)
2021	Screen Capture of Exhibit 2020 (YouTube) https://www.youtube.com/watch?v=d2an7LXhPSU
2022	Screen Capture of Exhibit 2020 (PacBio.com) https://www.pacb.com/videos/hifi-sequencing-see-what-youve-been-missing/
2023	Transcript of 05:05-06:06 from Exhibit 2020
2024	YouTube Video: "Methylation Detection with PacBio HiFi Sequencing: ASHG 2021." PacBio, October 27, 2021. URL: https://www.youtube.com/watch?v=rT23lBCp6Ro . Downloaded January 17, 2024.
2025	Screen Capture of Exhibit 2024 (YouTube) https://www.youtube.com/watch?v=rT23lBCp6Ro

Patent Owner's Exhibits	
No.	Description
2026	Transcript of 15:46–15:56 from Exhibit 2024
2027	Web page: PacBio Sequencing 101: The SMRT Cell in PacBio long-read sequencing. URL: https://www.pacb.com/blog/smrt-cell/ . Web page visited January 23, 2024
2028	<i>Take2 Technologies Limited v. Pacific Biosciences of California, Inc.</i> , 5:23-cv-04166-EJD, Dkt. 8-2, Exhibit D to the Complaint – PacBio SMRT® Link release notes (v11.0) (Complaint Ex. D)
2029	U.S. Patent Application Publication US2010/0221716 by Flusberg
2030	Declaration of Amy Candeloro, January 26, 2024
2031	Declaration of Chelsea Baker
2032	List of Materials Considered by Dr. Peter K. Koo
2033	Patent Owner's Notice of Deposition of Dr. Christophe Dessimoz, June 7, 2024
2034	Patent Owner's Notice of Deposition of Mozghan Novbakhtian, June 7, 2024
2035	Document entitled "MEASURING DNA METHYLATION WITH 5-BASE HIFI 16 SEQUENCING"
2036	LinkedIn Profile for Mozghan Novbakhtian
2037	Transcript of Deposition of Christophe Dessimoz, Ph.D., June 21, 2024
2038	Beaulaurier et al., "Deciphering bacterial epigenomes using modern sequencing technologies," <i>Nat Rev Genet</i> 20, 157–172 (2019) ("Beaulaurier")
2039	Birknerova et al., "Circulating Cell-Free DNA-Based Methylation Pattern in Saliva for Early Diagnosis of Head and Neck Cancer," <i>Cancers</i> , 14, 4882 (2022) ("Birknerova")

Patent Owner's Exhibits	
No.	Description
2040	Gouil et al., "Latest techniques to study DNA methylation," <i>Essays in Biochemistry</i> , 63, 639–648 (2019) ("Gouil")
2041	Supplemental Declaration of Dr. Peter K. Koo
2042	Sang, "Bioinformatics Analysis of DNA Methylation Through Bisulfite Sequencing Data," <i>Methods Mol. Biol.</i> , 2198:441-450 (2021) ("Sang")
2043	Wang et al., "Factors and Methods for the Detection of Gene Expression Regulation," <i>Biomolecules</i> , 13, 304 (2023) ("Wang")
2044	Cheung et al., "Direct haplotype-resolved 5-base HiFi sequencing for genome-wide profiling of hypermethylation outliers in a rare disease cohort," <i>Nature Communications</i> , 14, 3090 (2023) ("Cheung")
2045	Web page: Oxford Nanopore Technologies – How nanopore sequencing works. URL: https://nanoporetech.com/platform/technology . Web page visited July 25, 2024.
2046	Web page: Your Genome – What is Oxford Nanopore Technology (ONT) sequencing? URL: https://www.yourgenome.org/theme/what-is-oxford-nanopore-technology-ont-sequencing/ . Web page visited July 25, 2024.
2047	PDF downloaded from web page: Genome-wide CpG methylation calling with standard HiFi whole genome sequencing ("PacBio's Primrose Poster"). URL: https://www.pacb.com/wp-content/uploads/ESHG-2022-Genome-wide-CpG-methylation-calling-Blethrow-Poster.pdf . Web page visited July 25, 2024.
2048	Web page: ESHG Conference 2022. https://2022.eshg.org/ Web page visited July 25, 2024.
2049	Web page: ESHG 2022 Sequence Without Compromise PacBio. URL: https://programs.pacificbiosciences.com/l/1652/2022-03-17/43c8ch . Web page visited July 25, 2024.

Patent Owner's Exhibits	
No.	Description
2050	Searle et al., “Third-Generation Sequencing of Epigenetic DNA,” <i>Angew. Chem. Int. Ed.</i> 62, e202215704 (2023) (“Searle”)
2051	Gigante et al., “Using long-read sequencing to detect imprinted DNA methylation,” <i>Nucleic Acids Research</i> , 47(8), e46 (2019) (“Gigante”)
2052	CV of Dr. Peter K. Koo, updated July 26, 2024
2053	Web page: PacBio Transforms Access to the Epigenome and Streamlines Workflows, PacBio press release dated April 21, 2022. URL: https://www.pacb.com/press_releases/pacbio-transforms-access-to-the-epigenome-and-streamlines-workflows/ . Web page visited July 26, 2024.
2054	Comparison of '794 patent claim 1, EX1030, EX2024, and EX2047
2055	Additional list of Materials Considered by Dr. Peter K. Koo for preparation of Supplemental Declaration of Dr. Peter K. Koo (EX2041)
2056	Declaration of Amy Candeloro, July 26, 2024

I. INTRODUCTION

The Petition fails to demonstrate that claim 1 is obvious over the combination of Ground 1, the sole ground asserted such that patentability of claim 1 should be confirmed. Because the remaining claims 2-19 depend directly or indirectly from claim 1, the patentability of those claims should be confirmed for the same reasons. Claim 1 requires, in part, “receiving data acquired by measuring pulses in an optical signal” and from that data, “creating an input data structure” having a specific combination of features, as follows:

- (a) receiving data acquired by measuring pulses in an optical signal . . .;

- (b) creating an input data structure, the input data structure comprising a window of the nucleotides sequenced in the sample nucleic acid molecule, wherein the input data structure includes, for each nucleotide within the window, the properties:

- the identity of the nucleotide,

- a position of the nucleotide with respect to a target position within the window,

- the width of the pulse [PW] corresponding to the nucleotide, and

- the interpulse duration [IPD];

Ground 1 includes three of Petitioner’s own references: Flusberg, the ’338 patent and the PacBio-BAM. These references, however, do not disclose or

suggest the way in which claim 1 accounts for nucleotide identity, namely, creating an “input data structure” including “the identity of the nucleotide” with the other claimed properties. Instead, Petitioner’s expert testified that the Flusberg publications disclose a method that avoids including nucleotide identity in an input data structure as claimed and accounts for nucleotide identity by eliminating it as a variable. EX1002, ¶107 (“Flusberg’s specific example avoids using sequence context as an express input variable because it is held constant....”)

The Petition fails to provide motivation to modify the methods disclosed in Ground 1 that control for nucleotide identity by making it constant, with the method recited in claim 1 that requires that the “identity of the nucleotide” be included as a property in the input data structure. According to Petitioner’s expert, a POSA would understand that “[b]ecause the sequence context (e.g., identity of the bases in the vicinity of a modified base) strongly affects IPD and PW, it was further appreciated that context must be controlled for or explicitly evaluated when predicting base modifications.” EX1002, ¶ 35. In other words, the Petition asserts that a POSA would understand that the identity of the nucleotide must be accounted for in base modification detection. But, the purported need to account for nucleotide identity would not have motivated a POSA to modify the way nucleotide identity is accounted for in the Flusberg publications—eliminating it as a variable. Indeed, controlling for nucleotide identity as disclosed in the Flusberg

publications is not interchangeable with the method recited in claim 1 that includes nucleotide identity as a property in an input data structure.

Contrary to the assertion in the Petition, a POSA would understand that the well-characterized way to account for nucleotide identity in base modification detection at the time of the invention was to do exactly what the Flusberg publications disclose, control for nucleotide identity by keeping it constant. EX2041, ¶149 . In fact, the Petition cites a series of references that all follow the same path as the Flusberg publications, controlling for nucleotide identity, not including it as a property in an input data structure. EX2041, ¶135 *et seq.*

Moreover, objective indicia strongly favors that claim 1 is not obvious. For example, following publication of the invention Petitioner's own President and CEO praised the inventors of the '794 patent for their "development of a new method for highly accurate simultaneous determination of DNA sequence and CpG methylation in one go[]" that will "help researchers explore the impact of epigenetic changes in humans and other organisms and has great potential in clinical research and eventually diagnostics in cancer and other disease areas where methylation changes are known to be important disease markers." EX2017 at 5. After ten years of trying and failing to address the same long-felt need solved by the '794 patent, just ten months after Patent Owner published its invention, Petitioner announced its new method that now included nucleotide identity as a

property in an “input data structure” as recited in claim 1. EX2024. That Petitioner changed course from controlling for nucleotide identity and was able to introduce a new system incorporating Patent Owner’s invention just months after having access to the claimed method strongly favors patentability.

For the reasons set forth herein Patent Owner respectfully requests the patentability of claims 1-19 be confirmed.

II. THE ’794 PATENT

The ’794 patent discloses, *inter alia*, methods for determining nucleotide modifications, *e.g.*, methylation of DNA nucleotides, which is both essential for mammalian development and is also involved in a variety of biological functions and diseases. EX1001, 1:61-2:2. As such, the ’794 patent explains that “[t]he accurate measurement of methylomic modification on DNA molecules would have numerous clinical implications.” *Id.* 2:3-5.

The ’794 patent further explains the “bisulfite based approach” to modification detection has disadvantages including “that the bisulfite conversion step has been reported to significantly degrade the majority of the treated DNA”; “that the bisulfite conversion step would create strong CG biases” causing detection issues in the case of “DNA mixtures with heterogeneous methylation states”; and “bisulfite sequencing would not be able to sequence long DNA molecules because of the degradation of DNA during bisulfite treatment.”

EX1001, 2:19-28. (citations omitted)

So, the '794 Patent explains that as of its filing there was a “need to determine the modification of bases of nucleic acids, without prior chemical (e.g. bisulfite conversion) and nucleic acid amplification (e.g. using the PCR).” *Id.*, 2:28-31. “Achieving bisulfite-free determination of a base modification, include[ing] a methylated base, is the subject of different research efforts but none have been shown to be commercially viable.” *Id.*, 17:7-10.

The '794 patent solves that problem by disclosing “a new method that . . . allows the determination of base modifications, such as 5mC in nucleic acids without template DNA pre-treatment such as enzymatic and/or chemical conversions, or protein and/or antibody binding.” *Id.*, 2:35-39.

A. Claim 1

Claim 1 is directed to a “method for detecting a modification of a nucleotide in a nucleic acid molecule.” The method of claim 1 requires receiving a specific type of data as follows:

(a) receiving data acquired by measuring pulses in an optical signal corresponding to nucleotides sequenced in a sample nucleic acid molecule and obtaining, from the data, values for the following properties:

for each nucleotide:

an identity of the nucleotide,

a position of the nucleotide within the sample nucleic acid molecule,

a width of the pulse corresponding to the nucleotide, and

an interpulse duration representing a time between the pulse corresponding to the nucleotide and a pulse corresponding to a neighboring nucleotide;

Claim 1 further requires creating an “input data structure” in a particular format and including the combination of specific properties in that input data structure, as follows:

(b) creating an input data structure, the input data structure comprising a window of the nucleotides sequenced in the sample nucleic acid molecule, wherein the input data structure includes, for each nucleotide within the window, the properties:

the identity of the nucleotide,

a position of the nucleotide with respect to a target position within the window,

the width of the pulse corresponding to the nucleotide,

and

the interpulse duration;

In other words, claim 1 requires “receiving data,” then “obtaining, from the data, values for [several specific properties] properties,” and then “creating an input data structure” from that received data where for each nucleotide in the window, the “input data structure” includes “the identity of the nucleotide,” the

“position of the nucleotide with respect to a target position within the window,” the “width of the pulse corresponding to the nucleotide [PW]” and “the interpulse duration [IPD].” EX2041, ¶¶41-45.

Claim 1 further requires a model that is “trained by” “receiving a first plurality of first data structures” which have the same structure and properties as the “input data structure” as follows: “each first data structure comprising values for the same properties as the input data structure.” The “first data structures” are used to train the model as the claim explains “storing a plurality of first training samples, each including one of the first plurality of first data structures...” and “optimizing, using the plurality of first training samples, parameters of the model....” EX1001, 115:65-116:52; EX2041, ¶¶43-44. Both experts agree that a model should be trained by data structures that match the input data structure used for modification detection. EX2041, ¶44; EX1002, ¶130.

Finally, claim 1 step (d) recites “determining, using the model, whether the modification is present in a nucleotide at the target position....”

B. Prosecution of Claim 1

During prosecution, the examiner determined that the two primary references of Ground 1, Flusberg and the '338 patent, do not disclose or render obvious creating an input data structure having the following properties of claim 1, namely:

the input data structure comprising a window of the nucleotides sequenced in the sample nucleic acid molecule, wherein the input data structure includes, for each nucleotide within the window, the properties:

the identity of the nucleotide,

a position of the nucleotide with respect to a target position within the window,

the width of the pulse corresponding to the nucleotide,

and

the interpulse duration;

Specifically, following an interview discussing proposed claim amendments and both Flusberg and the '338 patent, the examiner stated that while “Flusberg displays various widths of pulses in their sequence data in a window,” Flusberg does not appear to “measure the widths as a data point to be combined with interpulse duration, nucleotide identity and position in determining the presence of mutations.” EX1004 at 217. The Examiner referred to that same reasoning for nonobviousness with respect to the '338 patent. *Id.* The full statement is as follows:

35 U.S.C. 102

Examiner stated the method of Flusberg displays various widths of pulses in their sequence data in a window, however it does not appear that they measure the widths as a data point to be combined with interpulse duration, nucleotide identity and position in determining the presence of mutations. Examiner will consider and compare future claim amendments with the prior art.

35 U.S.C. 103

See comments in 102 above.

EX1004 at 217.

The prosecution of the '794 patent further confirms the claimed “input data structure” is not simply “input data” or the output of SMRT sequencing, e.g., the Pac-Bio BAM.

Initially, the examiner rejected original claim 1 over both Flusberg and the '338 patent, asserting that the claimed “received” data, which in original claim 1 included the “input data structure,” was disclosed in both references. EX1004, 803-810. Original claim 1 recited, in part:

1. (Original) A method for detecting a modification of a nucleotide in a nucleic acid molecule, the method comprising:

receiving an input data structure, the input data structure corresponding to a window of nucleotides sequenced in a sample nucleic acid molecule, wherein the sample nucleic acid molecule is sequenced by measuring pulses in an optical signal corresponding to the nucleotides, the input data structure comprising values for the following properties:

for each nucleotide within the window:

an identity of the nucleotide,

a position of the nucleotide with respect to a target position within the respective window,

a width of the pulse corresponding to the nucleotide, and

an interpulse duration representing a time between the pulse corresponding to the nucleotide and a pulse corresponding to a neighboring nucleotide;

EX1004, 1090.

In response, Patent Owner amended original claim 1 to clarify that the claimed “input data structure” is not simply the data generated in the normal course of SMRT sequencing, e.g., the PacBio-BAM. Instead, the “input data structure” is created from “receiving data acquired by,” for example, PacBio’s SMRT sequencing. Specifically, claim 1 was amended to delete “receiving an input data structure,” and instead to recite “(a) receiving data acquired by...” and then “(b) creating an input data structure....” Amended claim 1 recited, in part:

1. (Currently Amended) A method for detecting a modification of a nucleotide in a nucleic acid molecule, the method comprising:

~~(a) receiving an input data structure, the input data structure corresponding to a window of nucleotides sequenced in a sample nucleic acid molecule, wherein the sample nucleic acid molecule is sequenced by receiving data acquired by measuring pulses in an optical signal corresponding to [[the]]nucleotides sequenced in a sample nucleic acid molecule, the input data structure comprising, and obtaining, from the data, values for the following properties:~~

~~for each nucleotide within the window:~~

~~an identity of the nucleotide,~~

~~a position of the nucleotide within the sample nucleic acid molecule with respect to a target position within the respective window,~~

~~a width of the pulse corresponding to the nucleotide, and~~

~~an interpulse duration representing a time between the pulse corresponding to the nucleotide and a pulse corresponding to a neighboring nucleotide;~~

~~(b) creating an input data structure, the input data structure comprising a window of the nucleotides sequenced in the sample nucleic acid molecule, wherein the input data structure includes, for each nucleotide within the window, the properties:~~

~~the identity of the nucleotide,~~

~~a position of the nucleotide with respect to a target position within the~~

~~window,~~

~~the width of the pulse corresponding to the nucleotide, and~~

~~the interpulse duration;~~

EX1004, 191.

The amendment confirms that simply having the data generated by SMRT sequencing, namely, the PacBio-BAM, does not disclose creating an “input data structure” having the claimed combination of features. Of course, the sequencing data was available in a computer readable format in both Flusberg and the ’338 patent. But, as the examiner recognized, the relevant question is whether Flusberg and the ’338 patent disclose or render obvious receiving that SMRT sequencing data and then creating an input data structure having claimed properties.

As explained above, the examiner determined that Flusberg and the ’338 patent do not disclose or render that combination obvious.

C. Claim Construction

Claims are construed using the claim construction standard under 35 U.S.C. 282(b), including construing the claim in accordance with the ordinary and customary meaning of such claim as understood by one of ordinary skill in the art and the prosecution history pertaining to the patent. 37 CFR § 42.100(b). That standard is applied herein.

D. Person of Ordinary Skill in the Art

For the purposes of this response, Patent Owner has applied Petitioner’s definition of a person of ordinary skill in the art (“POSA”). Paper 2 at 26; EX2041, ¶23.

III. CLAIM 1 IS NOT OBVIOUS OVER GROUND 1.

Obviousness requires consideration of the scope and content of the prior art,

differences between the prior art and the patent claims, the level of ordinary skill in the art, and any secondary considerations. *KSR Int'l Co. v. Teleflex Inc.*, 550 U.S. 398, 406 (2007), citing *Graham v. John Deere Co.*, 383 U.S. 1, 15-17 (1966). Obviousness must be considered based on the claim as a whole. *Ruiz v. A.B. Chance Co.*, 357 F.3d 1270, 1275 (Fed. Cir. 2004) (explaining that “[t]he ‘as a whole’ instruction in title 35 prevents evaluation of the invention part by part....”).

“In assessing the prior art, the PTAB ‘consider[s] whether a PHOSITA would have been motivated to combine the prior art to achieve the claimed invention.’” *In re Nuvasive*, 842 F.3d 1376, 1381 (Fed. Cir. 2016) (vacating and remanding PTAB decision finding claims unpatentable).

A. Flusberg, the '338 Patent and the PacBio-BAM do not Disclose or Render Obvious the Combination of Nucleotide Identity, Position, PW, IPD in an Input Data Structure of Claim 1.

The three references of Ground 1, Flusberg, the '338 patent, and the PacBio-BAM, do not render obvious the claimed “input data structure” including a window of nucleotides” and “for each nucleotide within the window” the claimed combination of “the identity of the nucleotide, a position of the nucleotide with respect to a target position within the window, the width of the pulse corresponding to the nucleotide, and the interpulse duration.” EX1001, claim 1.

As explained in the Institution Decision, “[r]elying on the testimony of Dr. Dessimoz, Petitioner argues that a person of ordinary skill in the art ‘would have

found it obvious to consider the claimed properties together because doing so represents nothing beyond the predictable use of known variables according to their well-characterized role in base modification detection.’” Paper 8 at 20 (citations omitted). But, according to Petitioner’s expert, the Flusberg publications “avoid” including identity of the nucleotide¹ as a property in an input data structure as required by claim 1. And the third reference, the PacBio-BAM does not disclose any method of detecting modified DNA and thus cannot remedy the deficiencies of the Flusberg publications.

In fact, Petitioner does not cite any reference in which the “identity of the nucleotide” generated during PacBio SMRT sequencing is included in an input data structure to predict base modification, let alone as a property included in an input data structure in combination with the other recited properties required by claim 1. EX2041, ¶135. So, contrary to Petitioner’s assertion that claim 1 is merely “the predictable use of known variables according to their well-characterized role in base modification detection,” if anything, the well-characterized role of nucleotide identity in base modification detection is to “avoid” including identity of the nucleotide as a property in “input data structure” by keeping the identity constant. EX2041, ¶149.

¹ “Identity of the nucleotide” is also referred to as “nucleotide identity.”

Similarly, at the time the '794 patent was first filed a POSA would not include “the width of the pulse corresponding to the nucleotide” in addition to the other properties in the claimed “input data structure.” EX2041, ¶151. As explained by several co-authors of the Flusberg publications, by 2019 a POSA would not include PW and instead focus on IPD. EX2041, ¶¶136-137.

Accordingly, because Petitioner has failed to demonstrate that claim 1 is obvious under Ground 1, Patent Owner requests that the patentability of claim 1 and its dependent claims be confirmed.

1. Identity of the nucleotide

The claimed “input data structure” requires, *inter alia*, “the identity of the nucleotide” in addition to several other properties. EX1001, claim 1. Yet, the Petition does not cite any reference disclosing a method where the “identity of the nucleotide” is included in an “input data structure” with any of the other properties of the claimed input data structure. Instead, the references cited by petitioner, including the references of Ground 1, disclose a method that “avoids” including the “identity of the nucleotide” as a property in an “input data structure” by holding the identity of the nucleotide constant. EX2041, ¶135. Thus, at best for petitioner, the “well-characterized role” of the “identity of the nucleotide” in the prior art, including all three references of Ground 1, is to avoid using it as a property in an input data structure as claimed by keeping it constant. *Id.*, ¶149.

Other than the purported “well-characterized role” of “the identity of the nucleotide,” the Petition provides no motivation for a POSA to modify the methods disclosed in the Ground 1 references to include the “identity of the nucleotide” in an input data structure in combination with the other recited properties. To the contrary, a POSA is not motivated to modify a method that is designed to avoid including the “identity of a nucleotide” as a property in an “input data structure” to instead include it. *Id.*, ¶132. The patentability of claim 1 should be confirmed for that reason alone.

According to Petitioner’s expert, a POSA would understand that “[b]ecause the sequence context (e.g., identity of the bases in the vicinity of a modified base) strongly affects IPD and PW, it was further appreciated that context must be controlled for or explicitly evaluated when predicting base modifications.” EX1002, ¶35. At deposition Dr. Dessimoz explained controlled for meant to “fix” the sequence context: “we cannot fix the pulse width or the interpulse duration. These are just observations. But we can fix the sequence context. And I think this is what they did . . . in the Flusberg publications.” EX2037, 87:19-23. Inexplicably, when asked, what “explicitly evaluated” meant, Dr. Dessimoz testified “I must admit, my answer is not very clean here. And I must confess, I don’t remember exactly what I meant by this.” *Id.*, 89:22-24. At best, all Dr. Dessimoz could make of his sworn statement was “we should not ignore the

sequence context.” *Id.*, 90:13-14. But, neither fixing nucleotide identity nor “not ignoring sequence context” would have motivated a POSA to modify the references of Ground 1 to include the “identity of the nucleotide” as a property in an input data structure with the other recited properties.

Moreover, Petitioner’s expert confirmed that the method disclosed in the Flusberg publications of controlling for nucleotide identity, is not interchangeable with the method recited in claim 1 that includes nucleotide identity as a property in an input data structure, as follows:

[Q:] Okay. Previously, we were talking earlier today, and you testified that sequence context needs to be taken into account, and we discussed two ways of doing it: The Flusberg method and the ’794 method. Do you recall that?

[A:] I believe I know what you're referring to. Yes.

[Q:] Would a person of skill in the art consider those two methods of taking into account sequence context interchangeable?

[A:] I mean, these are different kind of experiments, and the -- so, no, they cannot be viewed interchangeably. It's quite a different situation.

EX2037, 108:20-109:12.

Petitioner’s expert further testified that a POSA would not expect similar results from the two methods. EX2037, 109:18-24 (“So if taken as a whole, are you asking me about whether the results described in -- in the Flusberg publication, whether we might expect the same results in the ’794 experiments? Well, then my

answer would be no, because there are many differences between -- between the two.”) Petitioner’s expert later testified that a POSA would expect that the claimed method would work at least as well as the Flusberg methods. EX2037, 113:15-22.

Accordingly, even if the inclusion of the “identity of the nucleotide” as a property in an “input data structure” as claimed was a well-characterized use of nucleotide identity in base modification detection—it was not—a POSA would not have been motivated to modify the methods disclosed in the Ground 1 references that “avoid” including nucleotide identity as a property in an input data structure, with the claimed method. As the experts agree, the two approaches are not interchangeable. EX2041, ¶150; EX2037, 108:20-109:2.

a. No reference includes “identity of nucleotide” with any other property recited in claim 1.

First, no reference in Ground 1, includes identity of the nucleotide in an input data structure as claimed. Both Flusberg and the ’338 Patent provide methods that “control” for sequence context by eliminating nucleotide identity as a variable. EX2041, ¶88, 107, 127, 132. Specifically, Petitioner’s expert explained that Flusberg avoids including the identity of the nucleotide as a property in an input data structure as follows: “Flusberg’s specific example avoids using sequence context as an express input variable because it is held constant – Flusberg’s principal component analysis model is trained on a particular sequence that is varied in methylation state, but not in nucleotide sequence.” EX1002, ¶107.

Petitioner's expert further explained that "we can fix the sequence context. And I think this is what they did in this -- in this experiment reported in the Flusberg publications." EX2037, 87:21-23. In other words, the Flusberg publications fix nucleotide identity to avoid including it as a property in the "input data structure." Ex 2041, ¶132.

Indeed, as the Petition explains, "[t]he Flusberg Publications teach that pulse features (e.g., PW and IPD) are tabulated for a window of nucleotides and used in a previously trained model to determine whether a nucleotide is methylated." Paper 2 at 28. Accordingly, a POSA would understand that the methods of Flusberg are expressly configured to eliminate any need to further consider the identity of the nucleotides, and not to include the identity of the nucleotide in an input data structure as claimed in claim 1. EX2041, ¶¶149-50.

And, contrary to the assertion in the Petition at 29, the third reference of Ground 1, the PacBio-BAM, does not remedy the deficiency of Flusberg and the '338 Patent. EX2041, VIII.C. At deposition, Petitioner's expert explained that the reason he included the PacBio-BAM is to demonstrate that the required properties of the claimed "input data structure" were generated in SMRT sequencing and thus available for use in modification detection, not that the PacBio-BAM discloses what properties to use or how to use those properties in base modification detection, as follows: "I do not claim that [EX1011] teaches how to do base

modification detection. This is merely a description of the content of this file.”

EX2037, 105:22-24. Petitioner’s expert further explained the PacBio-BAM does not disclose how to use nucleotide identity as follows: “it is not a document that provides much information in terms of base modification detection.” *Id.*, 106:10-12.

In fact, the PacBio-BAM includes a single reference to “base modification detection” under the heading “[e]ncoding of kinetic pulse features,” which notes that two specific polymerase kinetic features, IPD and PW, the same two features used in the Flusberg publications, are preserved for “base modification detection.” EX1011 at 8. But, the PacBio BAM includes no reference to any specific method of detection or how to use any feature for base modification detection. EX 2041, ¶131. Moreover, a POSA would understand that “identity of the nucleotide” is not a “kinetic pulse feature.” *Id.* So, the PacBio-BAM does not suggest that the “identity of the nucleotide” should be used in modification detection. *Id.*

And, even if the PacBio-BAM did suggest that the “identity of the nucleotide” should be considered for base modification detection, the PacBio-BAM does not disclose or suggest how nucleotide identity should be used for base modification detection. It includes no disclosure of a method of detection. EX2037, 105:22-24. As such the PacBio-BAM does not motivate a POSA to modify the methods of Flusberg and the ’338 patent that fix nucleotide identity and

thus control for it, i.e., avoid including nucleotide identity in an “input data structure” as claimed, to arrive at the claimed method. EX2041, ¶131.

Moreover, contrary to Petitioner’s assertion, Patent Owner’s infringement contentions do not assert or admit that the PacBio-BAM is the claimed “input data structure.” Instead, the infringement contentions recite that the claimed “input data structure” having the claimed combination of features is “created” from the PacBio-BAM—not that it is the PacBio-BAM, as follows: PacBio-BAM is “used to create the data structure of claim element [1.b], i.e., an input data structure, for claim element [1.c].” EX1014, 25.

And, contrary to Petitioner’s expert’s assertion that the PacBio-BAM is an “off the shelf option” for the input data structure (EX1002, ¶ 82), a POSA would understand that while the PacBio-BAM includes information from which the input data structure could be created, the PacBio-BAM cannot be the claimed “input data structure” and does not suggest or provide motivation to select various data included in the PacBio-BAM to create the claimed input data structure. EX2041, ¶¶128-131.

In short, the PacBio BAM adds nothing to the disclosure of Flusberg or the ’338 patent. The Flusberg publications recognize that the identity of the nucleotide was generated during SMRT sequencing. But the methods disclosed in both Flusberg publications considered the identity of the nucleotide by controlling for it,

i.e., making it constant, thus avoiding including it as a property in an input data structure with other properties as recited in claim 1.

b. The Petition’s other listed reason do not support unpatentability under Ground 1.

First, Petitioner’s assertion that the PacBio-BAM “describes the file as compatible with ‘base modification detection’” (Paper 2 at 29) is incorrect and irrelevant. As explained above, the PacBio-BAM recites that two kinetic pulse features, IPD and PW, are encoded in the file in a manner that “[p]rovides a substantial disk-space savings without affecting important production use cases (base modification detection).” EX1011 at 8; EX2041, ¶131. A POSA would understand that disclosure in the PacBio-BAM simply indicates that those properties (IPD and PW) are preserved and available for use in base modification detection. *Id.* That disclosure does not teach or suggest a particular method of detection or that the file itself is an “input data structure” as claimed. *Id.*

Indeed, Petitioner’s expert clarified that the statement in his declaration that the PacBio-BAM is “a preexisting, off-the-shelf option for a data structure [and that a POSA could] use such a BAM file containing the recited properties as an input data structure” (EX1002, ¶ 82) meant that the PacBio-BAM included the information from which an input data structure could be created, as follows:

“Well, I think I used the words ‘off-the-shelf.’ And what I mean by that, it contains all the information needed, readily available, and it can be fed to the model without

needing to do any calculation. For instance, we do not need to go and retrieve additional information that would not be contained in the BAM file. It's just a straightforward operation to basically feed this information to however the model has been implemented.” EX2037, 102:10-20.

Petitioner's expert further clarified that when his declaration asserted that “person of ordinary skill in the art would have thus been motivated to use such a BAM file containing the recited properties as an input data structure, as expressly suggested in PacBio-BAM” (EX1002, ¶ 82), what he actually meant was that the PacBio-BAM only expressly refers to “IPD and PW” in relation to base modification detection (EX2037, 105:15-20), but that it must also include the identity of the nucleotide, because he was “hard pressed now to think of a use case of the BAM file that wouldn't involve the identity of the bases.” *Id.* at 105:11-13. But, just moments earlier Petitioner's expert testified that “in most contexts where you process a BAM file, you would want to read the identity of the base.” *Id.* at 105:1-2. “Most” does not mean “all.” Regardless, even if every use of a BAM file included using base identity, that base identity is used does not disclose, teach, or suggest how to use the identity of the nucleotide in base modification detection. EX2041, ¶131. As Petitioner's expert explained, “I do not claim that [the PacBio-BAM] teaches how to do base modification detection. This is merely a description of the content of this file.” *Id.* at 105:22-24. At best for Petitioner, the PacBio-

BAM suggests that the identity of the nucleotide should be considered in base modification detection. And, as explained above, the well-understood way to consider identity of the nucleotide in base modification detection in the prior art, included the Flusberg publications of Ground 1, was to control for it, and thus “avoid” including it as a property in the “input data structure” as recited in claim 1. EX2041, ¶142.

The Petition’s further assertion that kineticsTools (EX1021 at 1-2) is an example of “using PacBio’s BAM file as an input data structure for base modification detection” is incorrect. Petition, p. 29. At best as the Petition repeatedly acknowledge as does Petitioner’s expert, kineticsTools receives the PacBio-BAM file as an input, not as an input data structure. Petition, pp. 9, 22, 24, EX1002, ¶ 37 (“The kineticsTools manual describes a model for detecting modified bases that receives a PacBio BAM file as an input to the model...”).

kineticsTools is a tool provided by Petitioner to its customers that uses nucleotide identity in the same way as the Flusberg publications. Just like the Flusberg publications, kineticsTools “avoids” expressly considering nucleotide identity and instead controls for it by holding it constant. EX2041, ¶143; EX2037, 193:10-13 (“it is controlled in this case, and, yes, I think this is consistent with the method that is described in the white paper or in the Flusberg publications.”). As such, kineticsTools’ disclosure to “avoid” including nucleotide identity as a

property in an input data structure, cannot motivate a POSA to use the PacBio-BAM to modify the Flusberg publications that also “avoid” using nucleotide identity as recited in claim 1.

Moreover, kineticsTools does not use pulse width for base modification detection, let alone simply use the PacBio-BAM, which includes PW, as a input data structure for base modification detection. EX2037, 186:20-21 (“So based on my reading, it does not appear to be using pulse width.”); EX2041, ¶143. Thus, contrary to Petitioner’s assertion, the method disclosed in kineticsTools is not an example of “using PacBio’s BAM file as an input data structure for base modification detection.” Instead, just like the Flusberg publications, kineticsTools takes nucleotide identity into account by controlling for it, not including it in an input data structure as claimed. *Id.* And, kineticsTools further excludes use of pulse width for base modification detection in accordance with the understanding of a POSA by 2019. *Id.*, ¶151.

Next the Petition cites a string of references, including the Flusberg publications, that all follow the same path as the Flusberg publications explained above, namely, avoiding including identity of the nucleotide in “input data structure” as claimed by instead, controlling for it. EX2041, ¶134-43. In fact, Petitioner does not assert that any of those citations includes the identity of the nucleotide as a property in an input data structure as claimed. Again, contrary to

petitioner's purported proposition, the well-characterized role of the "identity of the nucleotide" in base modification detection, if anything, is to control for it by fixing or making it constant, in other words, avoid including it as a property in an "input data structure" as claimed. EX2041, ¶149-50.

Next, Petitioner's citation to Loomis (EX1009) as "corroborating the obviousness of considering data for nucleotide identity" does not disclose or suggest how to use nucleotide identity for base modification detection. Taking petitioner's statement as true, just like the Flusberg publications, Loomis simply suggests the obviousness of considering nucleotide identity, but does not disclose how nucleotide identity should be considered. EX2041, ¶138. In fact, just like the Petition, Loomis cites to Flusberg for "the previously described context effect on polymerase kinetics (Flusberg et al. 2010)." EX1009 at 126. Loomis then suggests that its "observations thus establish a clear foundation for the detection of epigenetic modifications within the CGG-repeat region, which has not been possible at the nucleotide level for full mutation alleles that are epigenetically silenced in FXS." EX1009 at 126. In other words, Loomis makes the same observation as the Flusberg publications and suggests that modification detection within the "CCG region" may be possible. EX2041, ¶138. But, Loomis does not disclose any method of detecting a modification or how nucleotide identity should be considered in such a method. *Id.* And, just like in kineticsTools, Loomis does

not use PW. *Id.*

Finally, Petitioner cites Liu (EX1022) as purportedly disclosing the combination of “both kinetic parameters and ordered nucleotide identities as an input data structure....” Paper 2 at 32. Petitioner concedes that Liu is directed to detecting base modification using a different type of DNA sequencing that has different sequencing data (“ONT data”). Paper 2 at 32. Contrary to the Petition, methods related to ONT data are not applicable to the polymerase kinetic data generating during SMRT sequencing. EX2041, ¶¶145-48.

First, claim 1 expressly excludes the data generated by Oxford Nanopore (“ONT”) sequencing, requiring “receiving data acquired by measuring pulses in an optical signal corresponding to nucleotides sequenced in a sample nucleic acid molecule.” EX1001, claim 1. ONT sequencing does not generate any optical signal data. Ex2041, ¶145. In contrast to PacBio SMRT sequencing that measures optical signals, ONT sequencing techniques measure electrical signals. *Id.* EX2037, 34:19-23. Liu further distinguishes PacBio SMRT sequencing from ONT sequencing. EX1022 at 2 (citing, *inter alia*, Flusberg (EX1006)). Other than determining nucleotide identity, ONT sequencing data does not correlate to PacBio SMRT sequencing data. EX2041, ¶¶145-48; EX2037, 33:14-34:4.

In fact, a fundamental basis for Petitioner’s expert’s opinion is that “[p]rior to the ’794 patent, persons of ordinary skill in the art appreciated that polymerase

kinetics were dependent on sequence context.” EX1002 ¶ 30. Polymerase kinetics are not found in ONT sequencing. As the Petition recites “SMRT sequencing obtained sequence and polymerase kinetic data by optically monitoring a single polymerase as it incorporates nucleotides into a growing complementary strand.” Paper 2 at 5. In contrast “ONT doesn’t use any DNA polymerases. Instead, it’s based on a small barrel-shaped protein in a membrane, called a nanopore, and measures changes to electrical current.” EX2046 at 2, 3. So, ONT sequencing does not include polymerase kinetics at all. *Id.* Not surprisingly, for ONT sequencing there is no relation between polymerase kinetics and sequence context. EX2041, ¶148. There are simply no polymerase kinetics in Oxford Nanopore sequencing. *Id.* As such, the other properties required in the input data structure of claim 1, namely PW and IPD which are both polymerase kinetics, are not found in ONT sequencing. As Petitioner’s expert testified, “I’m not making a claim here that there is a -- like, a one-to-one correspondence between every aspect of the electrical current to the -- to the optical signal.” EX. 2037, 240:6-9.

Petitioner’s expert’s creative quotation of Liu (EX1038) misrepresents the differences between PacBio SMRT sequencing and ONT sequencing with respect to “kinetics.” Petitioner’s expert claims that Liu discloses that “in each system, one can detect ‘modifications by measuring the kinetics,’ with only the specific kinetic parameters varying. . . .” EX1002, ¶41. But, the “quoted” sentence in Liu

actually reads as follows: “These technologies can detect RNA modifications by measuring the kinetics of the reverse transcriptase as it encounters a modified RNA -in the case of PacBio-, or by directly sequencing the RNA in its native form by pulling a native RNA through the nanopore -in the case of ONT-.” EX1038 at 2. The use of “by ..., or by ...” in that sentence makes clear that PacBio SMRT sequencing “can detect RNA modifications by measuring the kinetics of the reverse transcriptase as it encounters a modified RNA,” while ONT sequencing “can detect RNA modifications . . . by directly sequencing the RNA in its native form by pulling a native RNA through the nanopore.” *Id.* The citation does not support or imply that both systems can detect modifications by measuring kinetics. As set forth above, ONT sequence does not include polymerase at all, and therefore cannot include polymerase kinetics. EX2041, ¶148.

Thus, the use of nucleotide identity in Liu with ONT data does not disclose, teach or suggest the input data structure of claim 1 and does not motivate a POSA to modify the methods of the Flusberg publications that avoid nucleotide identity by controlling for it and instead include it as property in an “input data structure” as claimed. *Id.* The ONT data does not include two of the primary features of claim 1’s input data structure, IPD and PW. Moreover, the use of nucleotide identity with respect to a different type of sequencing that generates a different type of data disclosed in a single reference, cannot constitute Petitioner’s purported

“well-characterized use” of nucleotide identity in combination with the other recite properties of the claimed input data structure, properties that are only found in PacBio SMRT sequencing. *Id.*, ¶149.

For the foregoing reasons, a POSA would not be motivated to modify the methods of the Flusberg Publications to introduce a property that the Flusberg methods are designed to avoid. *Id.*, ¶142.

c. “Sequence context” does not mean “identity of the nucleotide”

A POSA would understand that the term “sequence context” as used in the art is not limited to “identity of nucleotide,” such that the need to consider sequence context in base modification detection does not disclose or teach, first, which sequence context properties should be considered, and second, how to consider any of those properties, let alone motivate a POSA to modify the Flusberg publications’ methods that avoid “identity of the nucleotide” and instead include the “identity of the nucleotide” as a property in an input data structure” as claimed. *Id.*, ¶149. Accordingly, the foundation for the Petition that a POSA’s purported understanding “that local sequence effects should be considered when predicting base modifications” (Paper 1 at 30) or that “sequence context (e.g., identity of the bases in the vicinity of a modified base) strongly affects IPD and PW, it was further appreciated that context must be controlled for or explicitly evaluated when predicting base modifications” (EX1002, ¶ 35) does not disclose suggest or

motivate a POSA to modify the methods of Flusberg and the '338 patent as claimed. EX2041, ¶149.

First, Petitioner's expert testified at deposition, that a POSA would understand that "sequence context" means "the signal and the identity of the bases that are surrounding a particular modification..." (EX2037, 80:19-20), and, that this is how the term is used in both Flusberg publications (*id.* at 83:11-22). Petitioner goes on to explain that "this context matters when it comes to the signal that is being measured, and in particular, the pulse width and the interpulse duration." In other words, according to Petitioner's expert "sequence context" refers to a window of information around a particular modified base, and includes for each position in that window both the signal, e.g., PW and IPD and other kinetic parameters, and the identity and position of the base in each of the positions within that window. Indeed, the Petition recognizes that the term "sequence context" includes more than just nucleotide identity and position. For example, in one passage, the Petition alleges that "POSAs appreciated that the sequence context (e.g., identity and positions of the bases in the vicinity of a modified base) strongly affects polymerase kinetics. . . ." Paper 2 at 30. The use of "e.g." confirms that sequence context refers to more than simply "identity and position of the bases." Accordingly, that "sequence context" should be considered does not teach a POSA which of those properties to use in base detection or how to use any

of one those properties. EX2041, ¶149.

Second, while recognizing that “sequence context” includes more than just identity and position, the Petition pulls a slight of hand by equating sequence context with nucleotide identity and position (see, e.g., Paper 2 at 2 (“sequence context (identity and position”) to claim that a POSA’s understanding that sequence context should be considered means not only that nucleotide identity should be considered, but that it should be considered in a particular manner, namely the claimed manner. But, even assuming that sequence context is synonymous with “identity of a nucleotide”—it is not—that a POSA understands that “sequence context” should be considered to detect a base modification, does not teach or suggest to a POSA how to consider the identity of the nucleotide. EX2041, ¶149. The references of Ground 1 avoid including nucleotide identity as a property in an input , data structure by controlling for it, as explained above. A POSA would not simply discard the method of Flusberg and the ’338 patent that controls for identity, and instead include nucleotide identity as property in a input data structure as claimed based on a disclosure that sequence content should be considered. *Id.*

2. A POSA would not have included pulse width with the other properties of the claimed input data structure.

Claim 1’s “input data structure” requires, *inter alia*, “pulse width” in addition to several other properties. At the time of the invention in 2019, the well-understood use of PW in base modification detection was to exclude it and instead

include IPD. EX2041, ¶151.

In 2012, PacBio released a White Paper on detecting DNA base modifications using SMRT sequencing. EX1016. (EX2037, 163:23-164:2). The White Paper describes two techniques for detecting DNA modifications both using IPD as the input: a comparison of IPD ratio between a DNA sample that is divided into native templates and whole- genome amplified control templates or a comparison between a native sample and an “*in silico* control [that] uses a predicted value of the mean IPD at each reference position that is based on the local sequence context.” EX1016 at 3. Fig. 4 of the White Paper illustrates the IPD kinetic signature detected by this method with IPD ratio plotted on the Y-axis v. template position on the X- axis. Notably, the White Paper omits pulse width as an input in either technique. EX2041, ¶142. No doubt the decision to select IPD as the input over pulse width or an IPD and pulse width combination is because that lack of sensitivity of pulse width to shifts in sequence context as the co-authors of Flusberg explained in 2013:

Given the high degree of regulation involved in the base incorporation process, PW is not as sensitive to shifts in sequence context induced by modifications as IPD (Flusberg et al. 2010). Therefore, for the purposes of developing an initial model of kinetic variation, we restrict attention to IPDs.

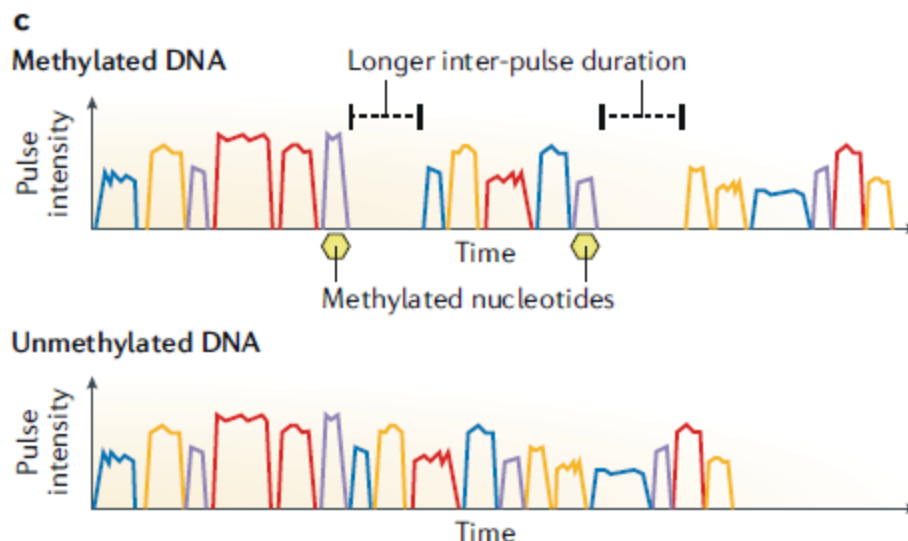
EX1010, 130. *See also* EX2041, ¶141.

Specifically, Schadt recites that Flusberg discloses “the possibility of directly observing chemical modification or damage events to nucleotides, and even discriminating between such events as a routine part of the SMRT DNA sequencing process.” EX1010, 130 (citations omitted). Schadt continues, “no statistical framework has been proposed to identify kinetic variation events from SMRT sequencing data that can serve as a surrogate for chemical modifications of DNA or RNA sequences.” EX1010, 130. In other words, according to some of the same authors of Flusberg and the ’338 patent, Flusberg discloses that it might be possible to detect modifications, but three years after Flusberg and the ’338 patent, no actual method of doing so had been proposed.

Then, in 2017, PacBio released kineticsTools. EX1021, EX2037, 164:3-8. And as Petitioner’s expert testified, kineticsTools illustrated “the state of the art.” *Id.* 189:18. Just like the White Paper, kineticsTools “loads IPDs observed at each position in the genome and compares those IPDs to values expected for unmodified DNA, and outputs the results of this statistical test.” EX1021, p.1. As in the White Paper, “[t]he expected IPD value for unmodified DNA can come from either an *in silico control* or an *amplified control*.” *Id.* Notably, kineticsTools released in 2017, did not use pulse width as an input to detect DNA modifications. EX2041, ¶143. “So based on my reading, it does not appear to be using pulse width.” EX2037, 186:20-21.

As set forth herein, Petitioner's Ground 1 does not rely upon Schadt or any one of several other publications by Petitioner attempting to solve the issue because Flusberg and the '338 patent directed a POSA down a different path, focusing on IPD as opposed to PW or identity of the nucleotide as a feature for an input data structure. EX2041, ¶151. Indeed, POSAs, and PacBio researchers and collaborators, followed that path until the '794 patent was first filed. EX2041 ¶¶136-143, 151.

And, a POSA and PacBio remained on the IPD path until the '794 patent showed the way to directly detecting DNA methylation, without bisulfite conversion, through SMRT sequencing. In 2019, just a few months before the '794 patent was filed, Schadt co-authored a paper titled *Deciphering bacterial epigenomes using modern sequencing technologies*. EX2038. In that paper, the Schadt authors explained how to detect DNA modifications using SMRT sequencing: "DNA modifications are detected by identifying changes in the kinetics of the polymerase as it translocates along the DNA template (FIG. 3c)." EX2038 at 159. Fig. 3c shows:



c | When observing the fluorescent traces produced by each ZMW, which are highly multiplexed on a chip, the order of pulses provides the read sequence, and pauses between pulses indicate the presence of a covalent modification in the template DNA.

EX2038 at 161.

The authors continued, “to detect modified nucleotides, IPD values from native DNA sequencing data can be compared with control IPD values from either methylation- free whole- genome amplified (WGA) DNA or precomputed in silico IPD models.” EX2038 at 159. “By comparing the native IPD values at a genomic position with either the predicted unmodified IPD value (using the in silico model) or a distribution of control IPD values obtained from WGA DNA, a simple statistical test can be used to identify methylated positions where the native and control IPD values diverge. *Id* at 160. In other words, the same techniques used in the White Paper and kineticsTools were still being used by PacBio to detect DNA

methylation with SMRT sequencing in 2019. PacBio continued down the IPD path, excluded the use of PW, and continued to control for sequence context.

Accordingly, at the time of the invention in 2019, the well-understood use of PW in base modification detection was to exclude it and instead include IPD.

EX2041, ¶151. Thus, a POSA would not have modified the Flusberg Publications to both include identity of the nucleotide, as explained above, and also both PW and IPD. By 2019, a POSA would have gone down a different path, focusing only on IPD.

IV. OBJECTIVE INDICIA OF NONOBVIOUSNESS SHOW THAT THE INPUT DATA STRUCTURE OF CLAIM 1 WAS NOT OBVIOUS AT THE TIME OF THE INVENTION.

An obviousness determination under § 103 requires consideration of objective indicia of nonobviousness. *WBIP LLC v. Kohler Co.*, 829 F.3d 1317, 1328 (Fed. Cir. 2016) (“[W]e have repeatedly stressed that objective considerations of non-obviousness must be considered in *every* case.”). Objective indicia of nonobviousness include commercial success, copying, industry praise, long-felt but unsolved need, failure of others, and unexpected results. *Volvo Penta of the Americas, LLC v. Brunswick Corp.*, 81 F.4th 1202, 1212 (Fed. Cir. 2023); *Power Integrations, Inc. v. Fairchild Semiconductor Intern., Inc.*, 711 F.3d 1348, 1368 (Fed. Cir. 2013). Because the “genius of invention is often a combination of known elements which in hindsight seem preordained,” objective indicia protect

against the prejudice of hindsight bias. *Power Integrations*, 711 F.3d at 1368.

Consequently, “objective indicia of nonobviousness are crucial in avoiding the trap of hindsight when reviewing, what otherwise seems like, a combination of known elements.” *Leo Pharm. Products, Ltd. v. Rea*, 726 F.3d 1346, 1358 (Fed. Cir. 2013).

There must be a nexus between the merits of the claimed invention and the objective evidence which can be shown one of two ways: 1) the patentee is accorded a presumption of nexus for objective indicia when the objective evidence is tied to a specific product and the product “embodies the claimed features” and “is coextensive with them” (*Henny Penny Corp. v. Frymaster LLC*, 938 F.3d 1324, 1332 (Fed. Cir. 2019)), or 2) the evidence of objective indicia is a direct result of the unique characteristics of the claimed invention (*Volvo Penta*, 81 F.4th at 1210). As shown below, here both are present.

A. The Method of Claim 1 Solved the Long-Felt Unsolved Need for Direct Detection of DNA Modifications by SMRT Sequencing Without Bisulfite Conversion.

A long-felt unsolved need evidences nonobviousness because it is reasonable to infer that the need would have not persisted had the solution been obvious. *WBIP, LLC*, 829 F.3d at 1332. It requires a showing that (1) the need was a persistent one recognized by those of ordinary skill in the art, (2) the long-felt need was not satisfied by another before the invention by the inventor, and (3)

the invention satisfies the long-felt need. *Id.* at 1332-1333.

1. The need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing was a persistent need recognized by a POSA.

Bisulfite sequencing treats DNA samples with bisulfite to convert unmethylated cytosines to uracil, leaving methylated cytosine intact. The treated DNA is PCR-amplified which converts the uracils to thymines. The amplified DNA can then be analyzed by microarray-based techniques or by 2nd generation DNA sequencing technologies, *e.g.*, Illumina sequencing. '338 patent, EX1007 at 2:28-39. EX2040 at 640. By the time the '794 patent was filed, a variety of bisulfite procedures had been developed. *See, e.g.*, EX2039 at 15, EX2038 at 158, EX2040 at 640.

Many human diseases are associated DNA methylation aberration, including carcinogenesis, imprinting disorders, repeat-instability disease, autoimmune disorders, metabolic disorders, neurological disorders and aging. EX1001, 1:61-2:2. "Changes in mC patterns have a crucial role in development and have been associated with cancer and other diseases." EX1006, at 461. Because the biological importance of 5-mC in mammals have been recognized for more than half a century, methylation detection has historically been devoted to 5mC. EX2038 at 158. As PacBio explains in its 2012 White Paper, methylation refers not any to any base modification, but rather 5mC. EX2016 at 1 ("Today, the most

commonly studied base modification is 5mC, commonly referred to as methylation despite the fact that other bases are also naturally modified by methyl groups.”).

a. Bisulfite sequencing was the most common DNA methylation detection method and the recognized gold standard.

From the time of the Flusberg publications through the filing of the '794 patent, bisulfite sequencing was the most common method used to detect methylated cytosine and widely recognized as “the gold standard” for single-nucleotide resolution methylation profiling and quantitative DNA methylation analysis, especially for characterizing 5mC. EX2041, ¶153; EX1007, 2:23-24 (“Bisulfite sequencing is the current method of choice for single-nucleotide resolution methylation profiling.”), EX1006 at 461 (“Currently, the most common technique for studying cytosine methylation involves bisulfite treatment...”); EX1016 at 1 (“Most high-throughput techniques focus only on cytosine methylation and involve both bisulfite sequencing to convert unmethylated cytosine nucleotides to uracil nucleotides...”); EX2038 at 158 (“the gold standard for characterizing 5mC in eukaryotic genomes (owing to its high sensitivity, accuracy and compatibility with NGS technologies), bisulfite sequencing...”), EX2040 at 640 (“These properties have made bisulfite converted DNA the gold standard of 5mC detection techniques;”); EX1001, 2:5-6 (“One widely used method to measure DNA methylation is through the use of bisulfite sequencing.”),

Ex 1030 at 1 (“The most widely used method for detecting 5mC involves bisulfite treatment...”), EX2042 at 441 (“[B]isulfite sequencing is widely used to determine the patterns of genomic methylation as a gold standard technology”); EX2039 at 15 (“Whole genome bisulfite sequencing (WGBS) is currently the gold standard DNA methylation profiling technology.”); EX2043 at 10 (“BS [bisulfite] sequencing and its derivative methods are gold standards for quantitative DNA methylation analysis.”).

Incredibly, PacBio’s expert has “only a superficial understanding” of bisulfite conversion and “was not so familiar” with some of the problems of bisulfite sequencing. EX2037, 141:18-19, 142:23-24.

b. The drawbacks of bisulfite conversion/sequencing created a persistent long-felt need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing.

Despite its gold standard status, bisulfite sequencing has significant drawbacks as Flusberg explains:

[T]here are several drawbacks to the technique. For example, the sample preparation steps associated with bisulfite treatment can be costly and time-consuming, and the harsh reaction conditions necessary for complete conversion can degrade DNA. Additionally, the reduction of complexity in converted genomes constrains primer design for subsequent PCR amplification and complicates alignment to a reference genome. Finally, discrimination between cytosine, mC and hmC cannot be accomplished with bisulfite treatment–based

sequencing.

EX1006 at 461. *See also* EX1007, 2:39-49, 68:62-69:2; EX2041, ¶153.

Flusberg '338 patent explains additional drawbacks with respect to *de novo* methylation profiling.

Furthermore, because bisulfite sequencing relies on either microarray or 2nd-generation DNA sequencing technologies for its readout of methylation status, it also suffers from the same limitations as do these methodologies. For array-based procedures, the reduction in sequence complexity caused by bisulfite conversion makes it difficult to design enough unique probes for genome-wide profiling. Most 2nd-generation DNA sequencing technologies employ short reads and thus have difficulties aligning to highly repetitive genomic regions. This is especially problematic, since many CpG islands reside in such regions. Given these limitations, bisulfite sequencing is also not well suited for *de novo* methylation profiling.

EX1007, 2:50-63 (citations omitted). *See also*, EX2041, ¶153

The first sentence of the Flusberg paper identifies the need: “the direct detection of DNA methylation, without bisulfite conversion, through single-molecule, real-time (SMRT) sequencing.” EX1006 at 461. PacBio’s expert, confirms that the Flusberg authors and the '338 patent inventors were attempting to solve that very need.

[Q.] [W]ouldn't you agree that both the paper and the patent are trying to use a PacBio sequencer to detect methylation without the

need for bisulfite conversion?

*

*

*

[A:] It certainly appears so based on the portion that you've recited.

EX2037, 145:9-15.

But, the Flusberg method failed to solve the need for the direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing because, as explained by Flusberg's co-authors in Schadt three years later "[p]resently, only 5-mC can be routinely detected using NGS technology and the only after the samples have been bisulfite treated, a procedure that creates a number of experimental and informatics challenges." EX1010 at 129; EX2041, ¶154. Schadt lamented that "no statistical framework has been proposed to identify kinetic variation events from SMRT sequencing data that can serve as a surrogate for chemical modifications of DNA or RNA." EX1010, at 130; EX2041, ¶155.

Notably, the three years between the Flusberg paper and the Schadt paper were "a period of rapid development" in a "very dynamic field," where "just a few years would already possibly make a difference in the amount of data that could be considered to train a model." EX2037, 157:9-14; 158:23-25, 178:25-179:1. Thus, Schadt's observation that bisulfite sequencing is still required for detecting DNA methylation and that SMRT sequencing cannot serve as a surrogate for chemical

(e.g., bisulfite) modifications reflects that notwithstanding the rapid development of this technology, the Flusberg methods were unable to solve the need for direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing.

And yet, nine years after Flusberg, the Lo inventors filed '794 patent directed to using SMRT sequencing to directly detect DNA methylation, without bisulfite conversion, through SMRT sequencing because the drawbacks of bisulfite sequencing (Ex. 1001, 2: 19-28) created a need to determine the modification of bases of nucleic acids, without prior chemical (e.g. bisulfite conversion) and nucleic acid amplification (e.g. using the PCR). *Id* at 28-31.

Solving that persistent need was the goal of Lo's '794 patent as Petitioner's expert testified: "The motivation to bypass bisulfite base approaches, yeah, I think this is indeed what is mentioned here in the background [of the '794 patent]." EX2037 206:12-19; EX2041, ¶59-60.

"The length of the time between the prior art publication date and the claimed invention can also qualify as an objective indicator of nonobviousness." *Leo Pharma*, 726 F.3d at 1359 (Fed. Cir. 2013). Here, nine years had passed since the Flusberg publications, a long time in a "a period of rapid development" in a "very dynamic field."

2. No one satisfied the long-felt need for direct detection of DNA methylation without bisulfite conversion through SMRT sequencing before the '794 patent.

“[L]ong-felt need is analyzed as of the date of an articulated identified problem and evidence of efforts to solve that problem.” *WBIP*, 829 F.3d 1334. Flusberg identified the need in 2010. Three years later when Schadt was published, the need had not been solved. For years, the PacBio scientists tried to solve the need for direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing to no avail.

During the years intervening between the Flusberg publications and '794 patent, PacBio developed kineticsTools for detecting DNA modifications without bisulfite conversion that used IPD, not PW or nucleotide identity as a property in an input data structure. kineticsTools did not solve the long-felt need as reflected by the publications of multiple authors, including PacBio authors, acknowledging the problems of direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing. EX2041, ¶157; EX2038 at 159 (“[D]etection of 5mC and 5hmC requires either high sequencing depth or additional steps to convert them to 5-formylcytosine and 5caC, both of which have higher signal-to-noise ratios.”); EX2040 at 643 (“[T]he subtle effect of 5mC and 5hmC increase the [strand coverage] requirements to 250X, unless they are enriched or modified to produce larger kinetic effect by glycosylation of TET-conversion to 5-

carboxylcytosine.^[2] PacBio sequencing therefore only achieves single-molecule resolution for certain marks and on relatively short fragments(≤ 2 kb) that can be read a large number of times by the polymerase.”); EX2051 (“We note that for the dominant eukaryotic genome base modification at 5mC, the PacBio technology requires very high coverage making it impractical for use in the analysis of mammalian genomes. PacBio SMRT sequencing can be combined with bisulfite treatment (SMRT-BS) to facilitate 5mc detection, but this approach is currently only available for targeted sequencing and the bisulfite treatment introduces the same drawbacks noted above in addition to fragmenting the DNA.”). In sum, at the time of the ’794 patent, the long-felt need for direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing and not been satisfied by others.

3. The input data structure of claim 1 directly detects DNA methylation without bisulfite conversion through SMRT sequencing and produces comparable results.

The ’794 patent tested various features as the input data structure and concluded that IPD, PW and nucleotide identify (sequence context) was the most

² TET-conversion is technically not bisulfite conversion, but it precludes direct detection of DNA methylation through SMRT sequencing because it requires enzymatic conversion of the DNA template prior to sequencing.

predictive feature set of DNA methylation detection. EX2041, ¶158.

We evaluated the power of each feature (sequence context, IPD, and PW) in predicting the methylation state of CpG by including a subset of the features in the model. In the training dataset, models with (i) sequence context only, (ii) IPD only, and (iii) PW only gave area-under-the-curve (AUC) values of 0.5, 0.74 and 0.86, respectively. While combining IPD and sequence context improved the performance with an AUC of 0.86. The combined analysis of sequence context ("Seq"), IPD, and PW substantially improved the performance with an AUC of 0.94 (FIG. 18A). The performance of an independent testing dataset was comparable to the training dataset (FIG. 18B).

EX1001, 32:18-29.

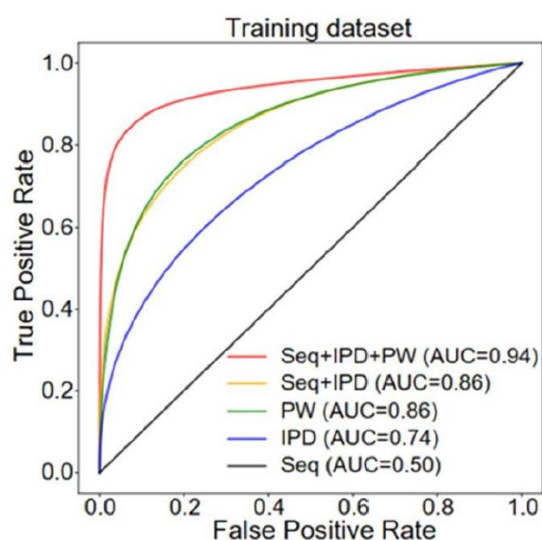


FIG. 18A

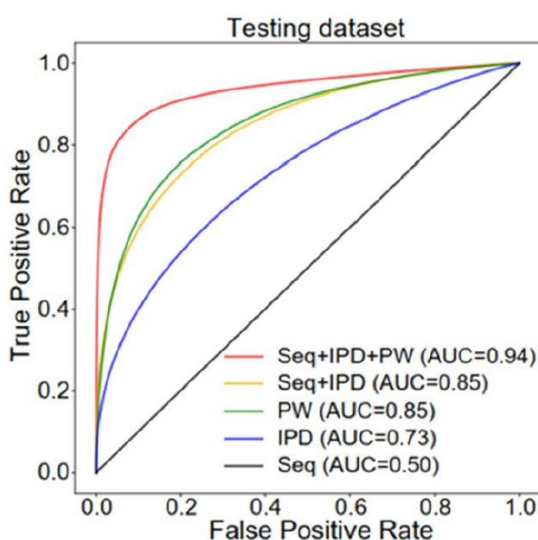


FIG. 18B

EX2001, FIGs. 18A, B.

Having determined that the identity of the nucleotide, the position of the nucleotide relative to a target position in a window, the PW corresponding to the nucleotide and the IPD were the best features for the input data set to detect DNA methylation, the Lo inventors trained their model according to the method of claim 1. EX2041, ¶158. Then, the Lo inventors conducted a series of experiments that compared the method of claim 1 to the “gold standard” – bisulfite sequencing. *Id.*

The '794 patent showed good correlation ($r = 0.90$ or higher) for most of the tested biological samples between bisulfite sequencing and the method of claim 1, which “demonstrated the feasibility for simultaneously analyzing copy number aberrations and methylation states for native molecules without any base conversions and amplifications prior to sequencing.” EX1001 at 48:55-63; EX2041, ¶¶158-59. “The embodiments disclosed herein have a number of exemplary advantages whereby it can be performed without bisulfite conversion that would degrade DNA drastically and can be performed without PCR amplification.” EX1001, 70:66-71:2.

For example, the '794 patent showed “a good correlation ($r: 0.96$, $p\text{-value}=0.0023$) between the overall methylation levels [in plasma DNA] deduced by the [claimed] approach and [conventional] bisulfite sequencing.” Ex1001 at 48:11-26; FIG. 51.

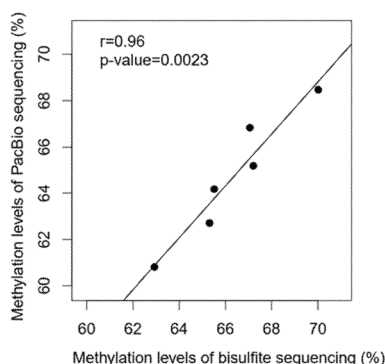


FIG. 51

As PacBio's expert recognized looking at this very data "But on the face of this good correlation, I believe this could be -- this could be qualified as a success, yes." EX2037, 207:16-208:4. And, as explained more in detail by Dr. Koo, the data disclosed in the '794 patent showed good correlation between the method of claim 1 and the gold standard - bisulfite sequencing for 5mC detection. In other words, the Lo inventors had solved the long-felt need for direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing. Similarly, Tse showed excellent correlation ($r = 0.99$, $P \text{ value} < 0.0001$) of overall methylation determination in 11 tissue DNA samples between bisulfite sequencing and the holistic model. EX1030 at 6, Fig. 4; EX2041, ¶158.

Additionally, the method of claim 1 significantly lowered the subread depth required for 5mC detection using SMRT without bisulfite conversion. EX2041, ¶159. PacBio's White Paper recommended a 250X minimum sequencing fold

coverage per strand for 5mC (EX1016 at 4) which even PacBio’s expert admits “is a high number” (EX2037, 184:4-5) so high that POSAs found the tool to shorter DNA fragments. EX2040 at 643.

B. PacBio Tried and Failed to Directly Detect DNA Methylation Without Bisulfite Conversion Through SMRT Sequencing

“There can be little better evidence negating an expectation of success than the actual reports of failure.” *In re Cyclobenzaprine Hydrochloride Extended Release Capsule Patent Litigation*, 678 F.3d 1063, 1081 (Fed. Cir. 2012). This is particularly true where others found development of the claimed invention difficult and failed to achieve any success, which can be *determinative* on the issue of obviousness. *Id.*

Flusberg tried to directly detect DNA methylation, without bisulfite conversion, through SMRT sequencing as the paper and PacBio’s expert confirmed. EX1006 at 461; EX2037, 145: 9-15. Yet, as Schadt explained, Flusberg failed to achieve that goal. For years the PacBio scientists tried to directly detect DNA methylation, without bisulfite conversion, through SMRT sequencing without success. EX2041, ¶157. PacBio’s “kineticTools” was not a viable solution because detection of 5mC and 5hmC required either high sequencing depth (250X) or additional steps to convert them to 5-formylcytosine and 5caC, TET conversion which is a chemical conversion similar to bisulfite conversion. *Id.* The high sequencing depth limits the fragment size to relatively

short fragments and is impractical for use in the analysis of mammalian genomes. EX2051, 2. In sum, at the time of the '794 patent, PacBio had tried and failed to directly detect DNA methylation, without bisulfite conversion, through SMRT sequencing.

C. The Holistic Kinetic Model Was Praised by PacBio and others in the field.

Industry praise of a product which embodies the patent claims weighs obviousness because industry participants are not likely to praise an obvious advance over the known art. *WBIP*, 829 F.3d at 1334; *Institut Pasteur & Universite Pierre Et Marie Curie v. Focarino*, 738 F.3d 1337, 1347 (Fed. Cir. 2013) (“[I]ndustry praise ... provides probative and cogent evidence that one of ordinary skill in the art would not have reasonably expected [the claimed invention].”).

The first praise came from Christian Henry, the President and CEO, who on February 10, 2021 “congratulate[d] Dr. Dennis Lo and colleagues at the Chinese University of Hong Kong on the development of a new method for highly accurate simultaneous determination of DNA sequence and CPG methylation in one go, published in the PNAS journal in January” in the Q4 2020 Earnings Call. EX2017 at 5. Mr. Henry emphasized the importance of Dr. Lo’s work, “[t]his new method will help researchers explore the impact of epigenetic changes in humans and other organisms and has great potential in clinical research and eventually diagnostics in

cancer and other disease areas where methylation changes are known to be important disease markers.” EX2017 at 5.

Mr. Henry praised “the method for highly accurate simultaneous determination of DNA sequence and CPG methylation in one go” which was published in the PNAS journal in January 2019. That paper, Tse et al., discloses the holistic kinetic model which Dr. Dessimoz agrees is the invention and corresponds to claim 1 of the ’794 patent. EX2037, 212:5-213:21. Dr. Koo agrees that the Tse discloses and is coextensive with claim 1 of the ’794 patent. EX2041, ¶¶77-87; EX2054. Just like *Henny Penny*, claim 1 is commensurate in scope of the product, the holistic kinetic model, and the “praise was generally directed to the claimed invention as a whole.” 938 F.3d at 1333.

The content of Mr. Henry’s praise in an earnings call is the type of industry praise that evidences an advance over the known art. Mr. Henry characterizes the Lo method as “new method” using PacBio’s SMRT sequencing “for highly accurate simultaneous determination of DNA sequence and CPG methylation in one go.” In “one go” reflects that no additional steps, such as bisulfite sequencing, are required. Mr. Henry recognized that Lo had solved the long felt need for direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing. While Mr. Henry’s motivation to praise the invention may have been to increase PacBio’s stock price and sell more sequencers, during an earning call

with Wall Street investors, it is highly unlikely that Mr. Henry would choose to highlight a trivial or nonobvious application of PacBio sequencing. He highlighted Dr. Lo's work because it was innovative, and he wanted to publicize the new method to improve PacBio's stock price and sell more sequencers.

PacBio's praise of the invention did not end there. In October 2021, at the 2021 annual meeting of the American Society of Human Genetics (2021 ASHG annual meeting), PacBio's President of Segment Marketing, Jennifer Stone, highlighted "the great work coming out of Dennis Lo's lab at the Chinese University of Hong Kong earlier this year." EX2020 (video) and EX2023 (transcript). Ms. Stone continued:

The team developed a methodology to directly examine 5-methyl-C at the whole genome level using the kinetic information from real-time HiFi sequencing without the need for bisulfite or enzymatic conversions. In the PNAS paper, Professor Lo et al. described the development of algorithms that leverage HiFi sequencing information from both DNA strands to improve the detection of 5-methyl-C. The paper describes its application to human whole-genome sequencing and the benefits of characterizing the epigenome with long accurate reads. It allows epigenetic phasing and allelic differential methylation. So just like the phasing of genetic variants, the ability to detect the methylation status genome-wide with long HiFi reads also allows for long-range epigenetic phasing information.

Id.

Others in the industry similarly praised the holistic kinetic model. In 2022, Birknerova et al. noted the persistent difficulty of detecting 5mC modifications with SMRT sequencing “[b]ecause the changes in the kinetic signal [IPD] caused by 5mC modifications[s] are extremely subtle, robust genome-wide measurement of 5mC modification is very challenging.” Yet, Birknerova et al. praised Tse’s holistic kinetic model as “highly correlated with bisulfite sequencing results, therefore the HK model constitutes a new, viable approach for studying epigenetic modifications in molecular diagnostic applications.” EX2039 at 16. “A new, viable approach for studying epigenetic modifications” reflects that Birknerova et al. considered the holistic kinetic model a nonobvious advance over the art.

Searle et al. summarized the difficulties PacBio authors had in direct detection of DNA methylation, without bisulfite conversion, through SMRT sequencing and remarked on the Tse paper as follows: “[r]ecently, a neural network was used to identify 5mC in native DNA, which significantly improved detection generating 99% agreement with bisulfite sequencing data.^[61] [61 is Tse] Uniquely, the neural network is able to simultaneously account for multiple variables of polymerase kinetics, including IPDs, pulse width and sequence context simultaneously, leading to greater accuracy.” EX2050 at 7. Searle et al.’s notation that the Tse showed 99% agreement with bisulfite sequencing data based upon multiple variables of polymerase kinetics reflects that the holistic kinetic model

was a nonobvious advance over the art.

D. PacBio Copied the Holistic Kinetic Model Disclosed in the Tse Paper.

Copying is an objective indicia of nonobviousness. *Liqwd, Inc. v. L'Oreal USA, Inc.*, 941 F.3d 1133, 1139 (Fed. Cir. 2019); *Crocs, Inc. v. International Trade Comm'n*, 598, 1294, 1311 (Fed. Cir. 2010) (“Copying may indeed be another form of flattering praise for inventive features.”); *Volvo Penta.*, 81 F.4th at 1213 (“[W]e have usually considered a determination of copying to be strong evidence of nonobviousness.”). “Copying requires duplication of features of the patentee’s work based on access to that work, lest all infringement be mistakenly treated as copying.” *Institut Pasteur & Universite Pierre Et Marie Curie v. Focarino*, 738 F.3d 1337, 1347-48 (Fed. Cir. 2013). Access to published articles about a patented method are relevant to the analysis of copying. *Institut*, 738 F3d at 1348.

Evidence of copying may include access and similarity to a patented product. *Medtronic*, 70 F.4th at 1340; *Liqwd*, 941 F.3d at 1337. Access coupled with circumstantial evidence regarding changes to a competitor’s design is sufficient to support copying. *DePuy Spine., Inc. v. Medtronic Sofamor Danek, Inc.*, 567 F.3d 1314, 1328-29 (Fed. Cir. 2009).

a. PacBio had access to the invention through the Tse paper

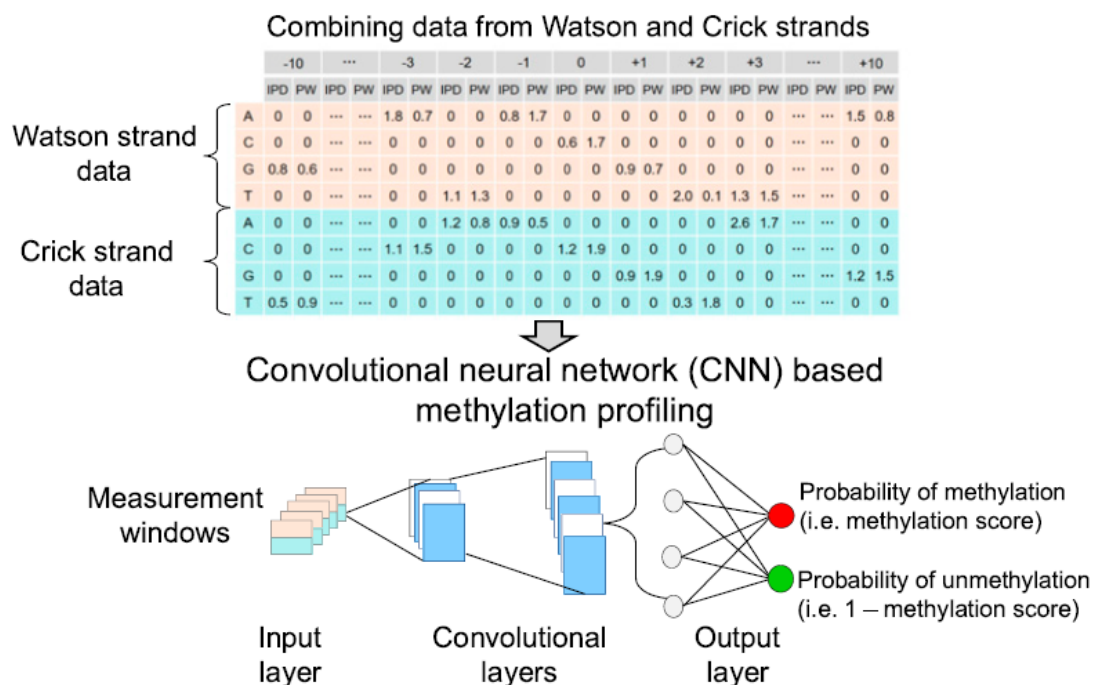
On January 25, 2021, Tse published the holistic kinetic model for DNA

modification detection. EX1030 at 1. As Dr. Dessimoz testified the holistic kinetic model disclosed in Tse is the invention of the '794 patent, claim 1. EX2037, 212:5-213:21. Dr. Koo agrees that the Tse discloses and is coextensive with claim 1 of the '794 patent. EX2041; ¶86; EX2054.

PacBio had access to the Tse paper as evidenced by the February 10, 2021 earnings call where Mr. Henry, congratulated Dr. Lo's team "on the development of a new method for highly accurate simultaneous determination of DNA sequence and CPG methylation in one go, published in the PNAS journal in January." Ex2017 at 5. The Tse paper (Ex. 1030) is the only PNAS published in January 2021 with Dennis Lo as a co-author. EX2041, ¶78.

b. The PacBio product bears a striking similarity to the invention disclosed in the TSE paper.

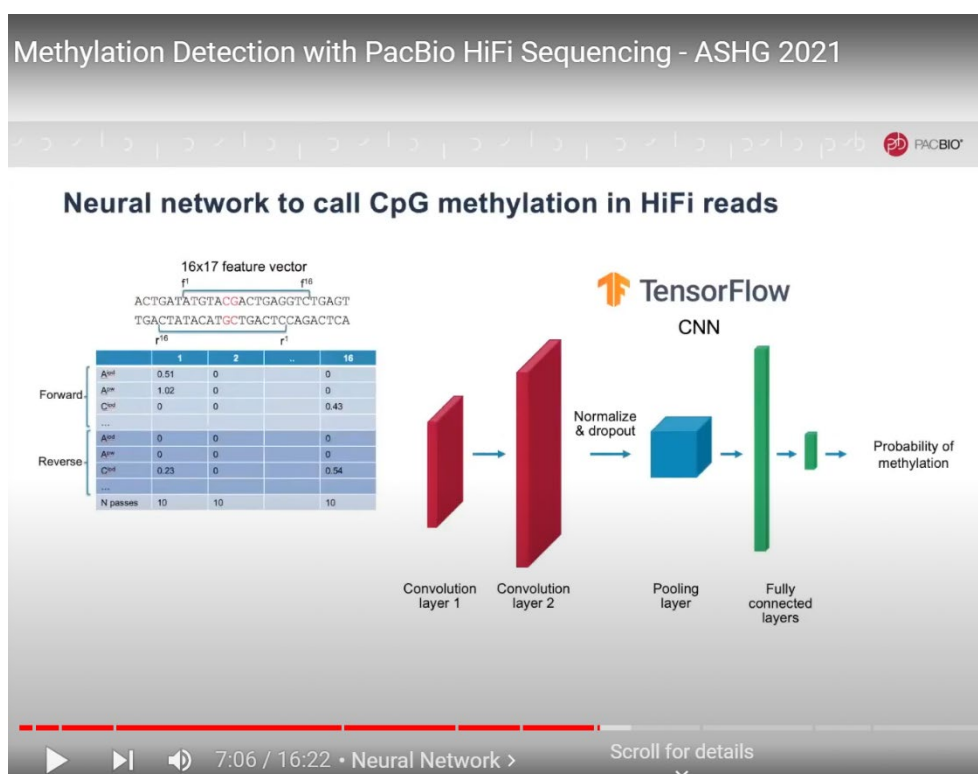
A portion of Figure 1 from Tse is set forth below. A matrix, titled "Combining data from Watson and Crick strands" depicts a 21 nucleotide window, 10 nucleotides upstream and 10 downstream of a cytosine within a CpG site. The nucleotides in the brackets were left out (denoted by ". . .") in the matrix. For a position of -3 corresponding to the base of adenine ("A"), the IPD (1.8) and PW (0.7) associated with "A" were filled in the corresponding cells between a column of "-3" and a row of "A." The other cells in the same columns were filled by "0." The other IPDs and PWs shown in the matrix were recorded based on the same rule. EX2041, ¶79-88; EX1030 at 2 (Fig. 1, Legend).



A number of combined Watson strand and the Crick strand measurement windows were used for training a CNN, so as to differentiate methylated and unmethylated cytosines in test samples. CNN involved input layer, convolutional layers, and output layer. The measurement windows were fed into the input layer, followed by the process of convolutional layers; then, the probability of methylation (range: 0 to 1) for a CpG was generated through the output layer based on a sigmoid function. This approach was referred to as the “holistic kinetic (HK) model” (HK model). EX2041, ¶79; EX1030 at 2 (Fig. 1, Legend).

While Jennifer Stone was praising Dr. Lo and his team’s work at the 2021 ASHG annual meeting, Aaron Wenger, PacBio’s Associate Director of Product Marketing presented “Methylation Detection with PacBio HiFi Sequencing” and announced that new “[t]ools available to call 5mC that are under development at

PacBio.” EX2024, 16:10-16:17. Dr. Wenger described the new tools as “models that integrate kinetic information over multiple bases and passes of a molecule” that “can provide high precision and recall for 5mC from PacBio HiFi reads.” *Id.*, 15:46-15:56. Dr. Wenger’s presentation showed a matrix as an input data structure for CNN modeling, trained by methylated and unmethylated samples, and accomplished 90% precision with 15-20 passes. *Id.* at 7:5-10, 14:58-end.



EX2024 screenshot at 7:06.

The above slide from Dr. Wegner’s presentation bears a striking similarity to the Fig. 1 of the Tse paper. EX2041, ¶¶80-81. The data matrix includes the kinetic data from the forward and reverse strands, i.e., the Watson and Crick strands. The matrix records the kinetic values (IPD and PW) around a window,

albeit, slightly shorter than the Tse window, here 16 nucleotides long. And, just like the Tse matrix, the Dr. Wegner's matrix records the IPD and PW for every nucleotide in the window. Here, the difference between the two matrices is that Tse uses two columns for each position, -10 IPD/ | PW...-3 IPD/PW, -2 IPD/PW, etc.) to record the IPD and PW values. EX2041, ¶81.

-10		...		-3		-2		-1		0		+1		+2		+3		...		+10	
IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW	IPD	PW

Whereas Dr. Wegner uses two rows A^{IPD} , A^{PW} , C^{IPD} , etc. As Dr. Koo notes, to the model, there is no distinction in the matrix format. EX2041, ¶82.

A^{IPD}
A^{PW}
C^{IPD}
...
A^{IPD}
A^{PW}
C^{IPD}
...
N passes

EX2024 screenshot at 7:06 (partial).

Dr. Wegner's matrix includes a row for the number of passes which refers to the subread depth. Tse also analyze the number of passes or subreads and found that "[t]he increase of subread depth would generally increase AUC values in differentiating methylated and unmethylated cytosines," which plateaued for the 21 nucleotide window at 30X. EX1030 at 5. EX2041, ¶84.

Dr. Wegner's slide indicates that the new tool uses a Tensor Flow

convolution neural network which is the comparable to the convolution neural network used in Tse. EX2041, ¶85. Dr. Wegner's slide shows convolutional layers as red rectangles which correspond to the blue and white rectangles in Tse Fig. 1. Dr. Wegner's slide includes a pooling layer, just like Tse. EX1030 at 10 ("A maximum pooling layer with a pool size of 2 was used."). And just like Tse, Dr. Wegner's slide includes connecting layers depicted as green rectangles which in Tse are white circles shown as connected to the green and red output layers. Lastly, Wegner's slide depicts a probability of methylation, just like the Tse Fig. 1. EX2041, ¶85.

c. After the TSE publication PacBio suddenly changed direction and adopted claim 1's input data structure.

A defendant's initial attempts at one design together with the prompt adoption of the claimed feature soon after the patent issued, are relevant indicia of nonobviousness. *Liqwd*, 941 F.3d at 1138; *Depuy*, 567 F.3d 1314, 1328 (Fed. Cir. 2009) ("Medtronic's engineers were focused on the same problem as the '678 patent....When the '678 patent issued the following month, however, Medtronic's team suddenly changed direction in the manner disclosed in the '678 patent."); *Dow Chem. Co. v. American Cyanamid Co.*, 816 F.2d 617, 622 (Fed.Cir.1987) ("Cyanamid tried but failed to develop the claimed invention and copied it instead.").

According to PacBio, the Flusberg publications in 2010 taught an input data

structure of PW and IPD as input variables to detect DNA modifications yet held sequence context constant. At that time, it was technically possible to practice the method of claim 1 with the claimed input data structure. EX2037, 136:16-25, 137:20-138:21. And, yet PacBio never did it. EX2037, 197:16-198:13, 200:17-201:6.

Because “PW is not as sensitive to shifts in sequence context induced by modifications as IPD,” PacBio eliminated PW from the input data structure and “restricted attention to IPDs.” EX1010 at 130; EX1016, at 3 (describes measuring “IPD distributions between a control sample and a native sample” to detect DNA modifications), EX1021 at 1 (“kineticsTool loads IPDs observed at each position in the genome, and compares those IPDs to value expected for unmodified DNA, and outputs the result of this statistical test”).

During the time between the Flusberg publications and the Tse publication, PacBio did not use PW or sequence context in a input data structure for DNA modification detection even though as Dr. Dessimoz admitted it was technically possible to do so. Tellingly, after the Tse publication PacBio reversed gears and adopted the claimed input data structure of Tse to detect DNA modifications.

Just nine months after the Tse publication, Dr. Wenger announced that new “[t]ools available to call 5mC that are under development at PacBio.” EX2024. Dr. Wenger’s presentation showed a matrix as an input data structure for CNN

modeling, trained by methylated and unmethylated samples, that accomplished 90% precision with 15-20 passes. *Id.* at 7:75- 10, 14:58-end.

The tools to call 5mC under development at PacBio, were apparently released in April 2022 as a PacBio press release announced that:

PacBio (NASDAQ: PACB), a leading developer of high-quality, highly accurate sequencing solutions, today announced the release of a transformative capability to detect DNA methylation using the Sequel IIe and Sequel II Systems. ***PacBio’s single-molecule approach provides a much more holistic view of molecular behavior during sequencing.*** Subtle patterns in this rich information allow detection of modified bases in native DNA during standard HiFi sequencing.

EX2053.

Tellingly, PacBio’s press release emphasizes the holistic view the purported “new” DNA modification detection method provides similar to the holistic kinetic model. Like the tool to call 5mC under development that Dr. Wenger present at the 2021 ASHG annual meeting, the release tool bears striking similarity to the holistic kinetic model. EX2041, ¶86.

Later, Dr. Wenger and other PacBio co-authors published a journal article Cheung et al. (EX2044) which outright admits that the holistic kinetic model of Tse was integrated into the PacBio DNA modification detection learning.

The technical ability to interrogate mCpG status from single

molecular real-time (SMRT) sequencing (kinetic) data was recently demonstrated using the HiFi-GS long-read platforms.²⁶ [Citation 26 is the Tse article Ex. 1030]. *This ability has now been integrated in the HiFi-GS production pipeline (Sequel IIe system release v11, PacBio, Menlo Park, CA)* that combines a deep learning model integrating sequencing kinetics and base context, to generate mCpG profiles genome-wide from standard sequencing libraries.

EX2044 at 2.

E. The Method of Claim 1 Had Unexpected Results Compared to Flusberg

When unexpected results are used as evidence of nonobviousness, the results must be shown to be unexpected compared to the closest prior art. *Millennium Pharmaceuticals, Inc. v. Sandoz, Inc.*, 862 F.3d 1356, 1368 (Fed. Cir. 2017).

“Unexpected results are useful to show the improved properties provided by the claimed invention are much greater than would have been predicted.” *Leo Pharma*, 726 F.3d at 1358. When a combination of known elements according to their established functions yields more than predictable results it is nonobvious. *Crocs, Inc. v. Int’l Trade Comm’n*, 598 F.3d 1294, 1309 (Fed. Cir. 2010).

“Consistent with the rule that all evidence of nonobviousness must be considered when assessing patentability, the PTO must consider comparative data in the specification in determining whether the claimed invention provides unexpected results. *In re Soni*, 54 F.3d 746, 750 (Fed. Cir. 1995).

Here the closest prior art are the Flusberg publications. EX2041, ¶160.

According to Petitioner's expert, the expectation of a POSA at the time of the invention would be that the method of the '794 patent, claim 1 would be comparable to the method of Flusberg.

My question is with the Flusberg publications in hand and the Flusberg methodologies in hand, what would a person of skill in the art think would be the result if instead of fixing the sequence context, as you referred to it, we used instead the input data structure of Lo? What would they have thought the result would be in 2019?

*

*

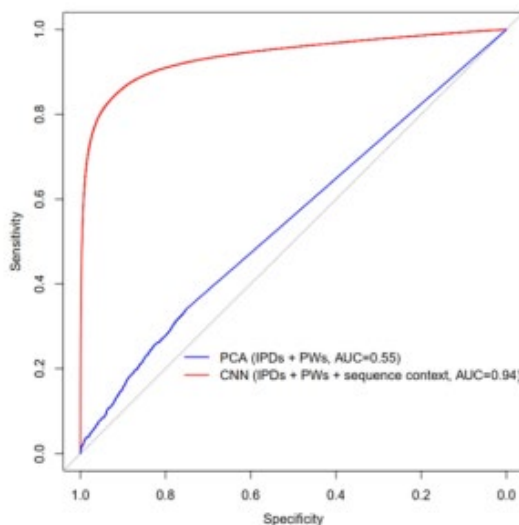
*

THE WITNESS: I think they would have had a reasonable expectation that Lo's methods would call -- successfully call modification of the basis because it follows very much the teaching that was known in the -- in the prior art records, including the importance of the context, the inter pulse duration, pulse width, and that entity of the basis. And, therefore, ***I think a person of ordinary skill in the art in 2019 would have expected, you know, Lo's approach to perform at least as well as the publication*** that was -- that came out nine years prior, using, you know, data that was necessarily much more rudimentary than what happened during this period. ***That would be the expectation. But, again, you know, the expectation is one thing. But usually a person of skill in the art runs the experiment, and this is what then provides a kind of more definitive indication.***

EX2037, 112:25-113105:10-114:1.

As Dr. Koo testified, a POSA could not predict the results of using the features of claim 1 of the '794 patent in light of the Flusberg publications, the results could be better or they could be worse, a POSA would need to run the experiment to determine the answer. EX2041, ¶161. Dr. Dessimoz essentially concedes this point when he says “expectation is one thing,” and goes on to admit, a POSA would not be able to predict the outcome until the experiment is run. Unpredictability of results equates more with non-obviousness than obviousness. *Honeywell, International, Inc. v. Mexichem Amanco Holdings S.A. DR C.V.*, 865 F.3d 1348, 1356 (Fed. Cir. 2017) (“[R]easoning that one would no more have expected failure than success is not a valid ground for holding an invention to have been obvious.”).

Yet, the comparative data in the '794 patent shows, however, that the method of claim 1 is far better than the method of Flusberg. EX2041, ¶162.



“FIG. 29 shows that the AUC of the method based on the principal component analysis for which two principal components were used in Flusberg (EX1006) involving IPDs and PWs.” EX1001, 35:38-44. In this instance, the ’794 patent claim 1 method produces an unexpected result relative to the closest prior art, Flusberg. The method of claim 1 results in a very high prediction (0.94) compared to the Flusberg method (0.55) which is about what would be expected by random guessing. EX2041, ¶164.

Additionally, as shown above, the method of claim 1 provides comparable results to bisulfite sequencing which is another unexpected result over Flusberg. EX2041, ¶165. *See also* EX2052, ¶87. And, while Flusberg predicted that someday in future *de novo* methylation profiling may become possible with SMRT sequencing. EX1006 at 465. With the input data structure of claim 1, the Lo inventors were able to successfully use SMRT sequencing for *de novo* methylation profiling of a variety of cells. EX2041, ¶165.

V. PATENTABILITY OF CLAIMS 2-19

Ground 1 is the sole ground asserted against claim 1. Because claim 1 is patentable over Ground 1 as explained above, and claims 2-19 all depend directly or indirectly from claim 1, the patentability of claims 2-19 should be confirmed for the same reasons as claim 1.

VI. CONCLUSION

For the foregoing reasons, Patent Owner requests patentability of the claims be confirmed.

Dated: July 26, 2024

Respectfully submitted,

/Michael J. Wise/

Michael J. Wise, Reg. No. 34,047
Lead Counsel

Perkins Coie LLP
1888 Century Park East, Suite 1700
Los Angeles, California 90067-1721
Phone: 310.788.9900
Fax: 310.788.3399

Back-Up Counsel
Joseph P. Hamilton, Reg. No. 51,770
Yingli Wang, Reg. No. 67,372

Attorneys for Patent Owner, The Chinese
University of Hong Kong

CERTIFICATE OF WORD COUNT UNDER 37 CFR § 42.24(d)

Under 37 C.F.R. § 42.24(d), the undersigned certifies that the word count for this Patent Owner's Preliminary Response to the Petition for *inter partes* review totals 13,817 excluding the parts exempted by 37 C.F.R. § 42.24(a). The word count was made using the built-in word count function in the Microsoft® Word software used to prepare this document.

Dated: July 26, 2024

Respectfully submitted,

/Michael J. Wise/

Michael J. Wise, Reg. No. 34,047
Lead Counsel

Perkins Coie LLP
888 Century Park East, Suite 1700
Los Angeles, California 90067-1721
Phone: 310.788.9900
Fax: 310.788.3399

Back-Up Counsel
Joseph P. Hamilton, Reg. No. 51,770
Yingli Wang, Reg. No. 67,372

Attorneys for Patent Owner, The Chinese
University of Hong Kong

CERTIFICATE OF SERVICE

Pursuant to 37 C.F.R. § 42.6(e), I certify that I caused to be served a true and correct copy of the foregoing: PATENT OWNER'S RESPONSE and accompanying exhibits by email to the electronic service addresses for Petitioner on the date indicated below:

LEAD COUNSEL	BACK-UP COUNSEL
Michael T. Rosato (Reg. No. 52,182) mrosato@wsgr.com Wilson Sonsini Goodrich & Rosati, 701 Fifth Avenue, Suite 5100, Seattle, WA 98104-7036 Tel.: 206-883-2529 Fax: 866-947-7329	Sonja R. Gerrard (Reg. No. 72,802) sgerrard@wsgr.com Wilson Sonsini Goodrich & Rosati, 701 Fifth Avenue, Suite 5100, Seattle, WA 98104-7036 Tel.: 206-883-2649 Fax: 866-947-7329
	Patrick M. Medley (Reg. No. 74,735) pmedley@wsgr.com Wilson Sonsini Goodrich & Rosati, 701 Fifth Avenue, Suite 5100, Seattle, WA 98104-7036 Tel.: 206-883-2593 Fax: 866-947-7329

Dated: July 26, 2024

Respectfully submitted,

/Amy Candeloro/

Amy Candeloro, Paralegal
Perkins Coie LLP