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Attorneys for *Plaintiff California Institute of Technology*

UNITED STATES DISTRICT COURT
NORTHERN DISTRICT OF CALIFORNIA
SAN JOSE DIVISION

CALIFORNIA INSTITUTE OF TECHNOLOGY,

Plaintiff,

v.

BIO-RAD LABORATORIES, INC.,

Defendant.

Case No.

COMPLAINT FOR PATENT INFRINGEMENT OF U.S. PATENT NO. 12,168,797

Plaintiff California Institute of Technology ("Plaintiff" or "Caltech"), by and through their counsel, for their Complaint against Defendant Bio-Rad Laboratories, Inc. ("Bio-Rad" or "Defendant"), alleges as follows:

NATURE OF THE ACTION

1. This is an action for infringement of U.S. Patent No. 12,168,797 (the "'797 Patent"), which is directed generally to certain systems, reaction mixtures, and kits for the multiplexed detection of analytes in a bio-chemical sample.

2. Traditional methods of detecting the presence or absence of multiple analytes in a bio-chemical sample involved, *inter alia*, using Polymerase Chain Reaction ("PCR") technology. These traditional methods have long been constrained by a fundamental limitation: each target must be tagged with a distinct fluorophore, which required tagging every target with a different color—*i.e.*, target A is tagged with a fluorophore that generates a "blue" fluorescence signal, target B is tagged with a different fluorophore that generates a "red" fluorescence signal, and so on. However, the number of distinct fluorophores that can be reliably detected by modern equipment is limited, significantly restricting the number of analytes that can be identified in a single test.

3. Beyond the limitation of fluorophore availability, traditional multiplexing also suffers from a problem known as degeneracy. Degeneracy arises when multiple analytes are tagged with the same fluorophore (*e.g.*, a first analyte is tagged with a "blue" fluorophore, a second analyte is tagged with a "red" fluorophore, and a third analyte is tagged with both "blue" and "red" fluorophores), leading to ambiguous fluorescence signals that do not uniquely identify a single analyte. In such cases, the system cannot distinguish between different analyte combinations, as multiple different scenarios could produce the same observed signal, making the results non-definitive (*e.g.*, detection of both "blue" and "red" fluorescence signals potentially identifies the presence of either the third analyte, both the first and second analytes, or all three analytes in the sample).

4. Dr. Axel Scherer, together with his colleagues at Caltech, revolutionized

multiplexing by eliminating both the one-target-one-color constraint and the degeneracy problem. Their innovation enables multiple targets to be unambiguously identified, even when they share the same fluorophore, by encoding detected signals using both fluorescence intensity and a decoding matrix, ensuring that each target produces a distinct, identifiable signal. This breakthrough significantly expands the number of detectable targets per reaction, improving the efficiency, accuracy, and cost-effectiveness of molecular diagnostics.

5. Plaintiff brings this action to stop Bio-Rad's infringement of this valuable and groundbreaking intellectual property. Bio-Rad markets and uses Caltech's patented multiplexing technologies without authorization and in violation of its exclusive rights.

6. As a result of Bio-Rad's infringement, and given the threat of its growing infringement, Plaintiff will be irreparably harmed if such infringement is not halted.

THE PARTIES

7. Plaintiff Caltech is a world-renowned science and engineering institute that marshals some of the world's brightest minds and most innovative tools to address fundamental scientific questions and pressing societal challenges. The mission of Caltech is to expand human knowledge and benefit society through research integrated with education. Caltech is an independent, privately supported institute located in Pasadena, California.

8. Defendant Bio-Rad is a Delaware corporation headquartered in Hercules, California. Bio-Rad is a global developer and manufacturer of products for the life science research and clinical diagnostics markets.

JURISDICTION AND VENUE

9. This Court has subject matter jurisdiction over this patent infringement action pursuant to 28 U.S.C. §§ 1331 and 1338(a).

10. This Court has personal jurisdiction over Bio-Rad because of Bio-Rad's

purposeful, systematic, and continuous contacts with California, and particularly the Northern District of California. Bio-Rad's headquarters are located in Hercules, California. Bio-Rad also sells products, including the infringing products (defined below), in the Northern District of California.

11. Bio-Rad conceded it is subject to personal jurisdiction in the Northern District of California when it moved to transfer another case adverse to Caltech to the this District. *See* Case No.: 2:23-cv-08417 (C.D. Cal), ECF 26, p. 11 (Bio-Rad's Motion to Transfer), in which Bio-Rad admitted that "Bio-Rad is thus subject to personal jurisdiction and venue is proper in N.D. Cal."

12. Venue is proper in the Northern District pursuant to 28 U.S.C. § 1400. The facts establishing this are included throughout this Complaint. Bio-Rad maintains regular and established places of business in this District, as identified above, and offers for sale and sells its infringing products in this District.

LITIGATION HISTORY BETWEEN THE PARTIES

13. Caltech owns multiple U.S. patents protecting its innovative multiplexing technologies, including U.S. Patent Nos. 10,068,051 (the "'051 Patent"), 10,770,170 (the "'170 Patent"), 11,827,921 (the "'921 Patent"), and the '797 Patent (collectively, the "HDPCR Patents"). These patents cover Caltech's groundbreaking high-definition PCR ("HDPCR") technology, which enables the unambiguous identification of more analytes (also referred to herein as "targets") than there are available fluorophore colors by implementing a novel encoding scheme based on both fluorescence color and intensity as determined from a cumulative fluorescence signal. Like the other HDPCR Patents, the '797 Patent claims priority to U.S. Provisional Application No. 61/594,480. The '797 Patent issued on December 17, 2024.

14. In 2012, a group of Caltech's researchers founded ChromaCode, Inc. ("ChromaCode"), a molecular diagnostics startup company with a focus on diagnostics and bioinformatics. On June 8, 2015, Caltech granted ChromaCode an exclusive license

to the HDPCR Patents and other patents for the purpose of developing and commercializing Caltech's patented multiplexing technologies.

15. Litigation involving Caltech, ChromaCode, and Bio-Rad over competing multiplexing technologies has been ongoing.

16. On September 20, 2023, following receipt of a letter in which Bio-Rad threated to enforce its patents against ChromaCode, ChromaCode filed a declaratory judgment action against Bio-Rad in this District, seeking a judgment of non-infringement regarding Bio-Rad's U.S. Patent Nos. 9,222,128 and 9,921,154 (collectively, the "Bio-Rad Patents"). *See ChromaCode, Inc. v. Bio-Rad Laboratories, Inc*., No. 5:23-cv-04823-EKL ("ChromaCode I").

17. Shortly thereafter, on October 5, 2023, ChromaCode filed a complaint in the Central District of California, alleging that Bio-Rad infringed certain Caltech and ChromaCode patents. *See ChromaCode, Inc. v. Bio-Rad Laboratories, Inc*., No. 2:23-cv-08417-RGK-PD ("ChromaCode II"). Caltech subsequently joined ChromaCode II as a Plaintiff. On December 7, 2023, the court transferred ChromaCode II to the Northern District of California, where it was reassigned as *ChromaCode, Inc. v. Bio-Rad Laboratories, Inc*., No. 5:23-cv-06360-EKL.

18. On August 14, 2024, the court consolidated ChromaCode I and ChromaCode II under *In re ChromaCode Litigation*, No. 5:23-cv-04823-EKL ("*In re ChromaCode Litigation"*), pursuant to Fed. R. Civ. P. 42(a), recognizing the substantial factual and legal overlap between the cases.

19. During discovery in the *In re ChromaCode Litigation*, Caltech produced the '797 Patent to Bio-Rad on December 21, 2024, and Caltech expressly notified Bio-Rad's counsel that the patent could be reviewed and shared with Bio-Rad.

20. The *In re ChromaCode Litigation* has already progressed significantly, with extensive discovery—including depositions and the exchange of millions of pages of electronically stored information ("ESI"). The Court held a Markman hearing on

December 12, 2024.

21. Given the advanced stage of the *In re ChromaCode Litigation*, the present action should proceed independently to avoid delays and promote judicial efficiency. Also, whereas ChromaCode is a party to the *In re ChromaCode Litigation*, it is not a party to the present dispute, is not an exclusive licensee of the '797 patent, and no longer has exclusive rights to any of the patents in the '797 patent's family.

BACKGROUND

22. The patented technology at issue in this case concerns the ability to unambiguously detect multiple targets in a single sample using fewer color channels than targets. This novel approach offers a fast and inexpensive alternative to researchers and clinicians who need to unambiguously identify a large number of genetic targets in a single test.

A. Traditional Methods of Identifying Multiple Targets in One Sample

23. For years, researchers invested substantial resources and time to increase the number of targets that could be identified in a single sample using fluorescence-based detection. Early approaches relied on a **one-target-per-color system**, where each target was assigned a distinct fluorophore. In these methods, fluorescent probes were designed to bind to specific targets. When excited, these probes emitted light at different wavelengths (each corresponding to a color), and detectors measured the presence or absence of each respective target based on its fluorescence signal. However, the broad emission spectra of standard fluorophores often overlap, leading to spectral bleed—where signals from different fluorophores interfere with each other. This spectral overlap limits the number of analytes that can be simultaneously detected to the number of spectrally resolvable fluorophores. Consequently, increasing the number of detectable analytes necessitates the development or identification of additional fluorophores with minimal overlapping spectra, which is both challenging and impractical. For example, PCR systems today generally operate with one, two, four, or six available color channels.

24. To bypass the need for additional spectrally distinct fluorophores, researchers explored encoding analyte detection using a single fluorophore to detect multiple analytes. However, this approach led to a new fundamental limitation—**degeneracy**. Degeneracy occurs when multiple fluorescent probes share the same fluorophore, leading to fluorescence signals that do not uniquely correspond to a single target. Instead, different target combinations can generate the same measured fluorescence output, making it impossible to determine which specific analytes are present. For example, if two different targets (A and B) use the same fluorophore, and the detector measures fluorescence at that wavelength, the system cannot distinguish whether only A is present, only B is present, or both A and B are present. This ambiguity severely restricts multiplexing capabilities and introduces uncertainty in diagnostic assays.

25. To address the degeneracy problem, researchers combined additional labor-intensive processing steps along with fluorescence to identify more targets from a single sample, including aliquoting, spatial arraying, and sequential processing. These additional processing steps are labor-intensive and frequently require relatively expensive and complex optical and mechanical systems, and often times rely on statistical analysis to estimate the presence or absence of individual targets in a sample. Although these tools offered some ability to increase the number of targets detected in a sample, there remained a significant need for inexpensive multiplexed encoding and decoding methods that could identify unambiguously and deterministically a significantly higher number of targets.

B. Caltech Broke the Degeneracy Barrier

26. Recognizing the limitations of traditional fluorescence-based multiplexing,

researchers at Caltech—including Drs. Emil Kartalov,[1] Aditya Rajagopal,[2] and Axel Scherer,[3] developed a breakthrough multiplexing technology that eliminated the

[1] Dr. Emil Kartalov earned his B.S. in Physics (1998), M.S. in Applied Physics (2004), and Ph.D. in Applied Physics (2004), all from Caltech. His doctoral thesis focuses on single-molecule fluorescence microscopy, single-molecule DNA sequencing, and microfluidic DNA sequencing by synthesis. The first two developed techniques resulted in the founding of Helicos Biosciences in Boston, MA. For his postdoctoral work, Dr. Kartalov moved to the Biochemistry Department and later the Pathology Department at the Keck School of Medicine at the University of Southern California ("USC"), where his focus shifted to fundamental microfluidics and biomedical applications of microfluidic devices. Dr. Kartalov invented microfluidic vias and resultant autoregulatory devices and built a multi-analyte microfluidic immunoassay chip for protein diagnostics in fingerprick amounts of human serum and plasma. In 2006, Dr. Kartalov won the NIH K99/R00 Career Award and became tenure-track faculty in Pathology at USC in 2008. His group focuses on microfluidic point-of-care diagnostics and techniques for high-throughput analysis of pathology tissue slices. Dr. Kartalov has over 18 peer-reviewed publications and 11 issued patents. In 2016, Dr. Kartalov departed USC, and he is currently an Associate Professor at the Naval Postgraduate Institute.

[2] Dr. Aditya Rajagopal earned his B.S. (2008), M.S. (2010), and Ph.D. (2014) in Electrical Engineering, all from Caltech. Dr. Rajagopal's research in medical engineering, microfluidics, nanotechnology and bioengineering has resulted in numerous innovative technological developments. He is the inventor of core technologies related to not only multiplexed PCR assay design, but also combinatorics for biochemical labeling and algorithms for data processing. In the aggregate, Dr. Rajagopal has authored over 45 patents and patent publications. He is the recipient of numerous honors, including the Caltech Grubstake Award in 2017, the Caltech Innovation Initiative (CI2) in 2014 and 2015, the Demitriadis-Kafka-Kokallis Thesis Prize in January 2013, and various fellowships and scholarships sponsored by the National Science Foundation and Carnation Foundation from 2006 to 2013. In addition to co-founding ChromaCode, Dr. Rajagopal co-founded a healthcare company called Esperto Medical—a venture-backed Caltech spin-out utilizing compressed sensing methods with ultrasound to continuously and non-invasively measure patient blood pressure.

[3] Dr. Axel Scherer obtained his Ph.D. from the New Mexico Institute of Mining and Technology. He is the Bernard Neches Professor of Electrical Engineering, Physics, and Applied Physics at Caltech, where his research focuses on the design and microfabrication of optical, magnetic and fluidic devices. He is also a distinguished visiting professor at Thayer School of Engineering at Dartmouth College. He is known for fabricating the world's first semiconducting vertical-cavity surface-emitting laser ("VCSEL") at Bell Labs, now widely used in data communications systems. More recently, his group developed electromagnetic design tools and fabrication techniques for the definition of lithographically integrated optical devices. This led to pioneering work in photonic bandgap lasers, silicon photonic circuits, as well as tunable microfluidic dye lasers, leading to new classes of integrated optics. The first demonstration of strong coupling between single quantum dots and optical nanocavities recently emerged from a collaboration between Dr. Scherer and Dr. Hyatt Gibbs. Collaborations with Dr. Larry Dalton (University of Washington) resulted in some of the world's smallest and fastest light modulators. Dr. Scherer also fabricated some of the first surface plasmon enhanced high brightness light emitting diodes. His group miniaturized fluidic systems and demonstrated the first multi-layer replication molded fluidic chips, with

degeneracy problem and broke the one-target-per-color constraint. The solution enabled high-definition PCR multiplexing—specifically, the unambiguous identification of more targets from a single sample reaction than there are available color channels in the PCR instrument. As a result of continued diligent research efforts, the Caltech researchers' high-definition multiplexing solution was reduced to practice by at least February 3, 2012, when Caltech filed U.S. Provisional Patent Application No. 61/594,480, to which the '797 Patent claims priority.

27. Caltech's innovative approach to high-definition multiplexing, referred to as HDPCR[4], eliminates degeneracy by encoding analyte detection using cumulative signal intensities and a decoding matrix rather than relying solely on color presence. Instead of treating fluorescence as a binary presence or absence measurement, HDPCR measures fluorescence intensity at each channel. This ensures that even if multiple analytes share the same fluorophore, their unique cumulative fluorescence intensity signatures allow them to be distinguished. By setting up the PCR reaction and fluorophore "probe" mix in accordance with a predetermined encoding matrix, each target produces a distinct cumulative fluorescence intensity signature that can be uniquely decoded, meaning that even when multiple targets share the same fluorophore, the cumulative fluorescence intensity is different from that of a single target alone. This prevents different target configurations from producing identical fluorescence outputs, eliminating degeneracy. A

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thousands of valves creating microfluidic "laboratories" and single cell analysis systems. He leads a group focused on the miniaturization and integration of fluidic, optical, electronic and magnetic devices for applications in biotechnology. Dr. Scherer has co-authored over 300 patent publications and holds over 100 patents on the area of microfabrication and design of devices.

[4] "PCR" stands for the Polymerase Chain Reaction, which is a method of exponential amplification of specific target DNA in a reaction mix with a DNA polymerase and primers. Primers are short single-stranded DNA oligonucleotides, which bind to particular regions of a target sequence. The reaction mixture is cycled in heating and cooling steps. The heating cycle denatures or splits the double-stranded DNA target into single-stranded templates. In the cooling cycle, the primers bind to the target. After the template is primed, the DNA polymerase creates a copy of the original template. Repeated cycling exponentially amplifies the target 2-fold, with each cycle leading to approximately a billion-fold increase of the target sequence in 30 cycles.

processor-controlled analyzer applies a mathematical decoding matrix to the detected fluorescence signals. This allows the system to unambiguously identify each target, even when multiple targets fluoresce at the same wavelength. Because the decoding matrix assigns a unique intensity signature to each target, the system avoids misinterpretations.

28. The commercial impact of HDPCR is significant, as it enables the development of faster, more cost-effective, and minimally invasive PCR assays that can detect a wide range of diseases and medical conditions. Caltech's patented HDPCR technology directly addresses a critical industry gap, providing a scalable solution for high-throughput, highly accurate molecular diagnostics.

29. The global PCR market is immense. In the United States alone, the PCR market as of 2023 was 40% of the global $9.15 billion total market size, and is estimated to grow to $6.82 billion by 2032. *See* https://www.fortunebusinessinsights.com/polymerase-chain-reaction-pcr-market-102528 (last visited on February 18, 2025). The increase in PCR research and forensic laboratories and increasing demand for advanced diagnostics, which depend on the ability to detect multiple target sequences in single PCR reactions, are expected to drive market growth. Among other things, the increase in the prevalence of chronic and infectious diseases and genetic disorders is expected to boost PCR demand.

30. Bio-Rad's infringement of the '797 Patent has caused, and if not enjoined, will continue to cause significant economic harm to Caltech, including by diminishing the value of Caltech's intellectual property portfolio. Caltech's patents are among its most valuable assets, and its ability to license and protect its proprietary innovations is essential to maintaining its standing as a leader in scientific research and development. By infringing the '797 Patent, Bio-Rad has undermined the exclusivity and commercial value of Caltech's intellectual property, weakening its ability to negotiate fair licensing agreements and diminishing the overall worth of its patent portfolio.

31. Caltech will also suffer irreparable harm if Bio-Rad is not enjoined from

selling its infringing products and further infringement of the '797 Patent. Caltech's primary asset is its intellectual property, and its value is inextricably linked to its ability to protect and enforce its patent rights. If Bio-Rad is permitted to continue its unauthorized use of Caltech's patented technology, it will irreparably reduce the market value of Caltech's innovations, disrupt licensing opportunities, and weaken Caltech's ability to continue investing in groundbreaking scientific research. Even if Caltech recovers monetary damages from Bio-Rad, the harm caused by Bio-Rad's infringement cannot be fully remedied through financial compensation alone.

THE ASSERTED PATENT

A. The '797 Patent

32. On December 17, 2024, the USPTO issued the '797 Patent, entitled "Signal Encoding and Decoding in Multiplexed Biochemical Assays," to Drs. Emil Kartalov, Aditya Rajagopal and Axel Scherer. Caltech is the assignee of the '797 Patent. A true and correct copy of the '797 Patent is attached hereto as **Exhibit A**.

33. The '797 Patent claims priority to U.S. Provisional Application No. 61/594,480, filed on February 3, 2012.

34. The '797 Patent has 4 independent claims, claims 1, 19, 22, and 30. Claims 1 and 19 are directed to a system for detecting multiple analytes using a multi-channel detection system and a processor-controlled analyzer. Claim 22 is directed to a composition of matter, specifically a reaction mixture containing hybridization probes conjugated to fluorophores for multiplexed detection. Claim 30 is directed to a kit comprising the components necessary for unambiguous polynucleotide analyte detection, including the reaction mixture and instructions for its use.

35. Claim 1 of the '797 Patent recites:

> 1. A system comprising:
> a sample chamber configured to house a sample and analyte-specific reagent mixtures of analyte-specific hybridization probes and multiple fluorophores;
> a multi-channel detector to detect:

a first electromagnetic signal at a first wavelength from the sample chamber, the first electromagnetic signal generated by excitement of a first fluorophore of the multiple fluorophores;
a second electromagnetic signal at a second wavelength from the sample chamber, the second electromagnetic signal generated by excitement of a second fluorophore of the multiple fluorophores;
a third electromagnetic signal at a third wavelength from the sample chamber, the third electromagnetic signal generated by excitement of a third fluorophore of the multiple fluorophores;
a fourth electromagnetic signal at a fourth wavelength from the sample chamber, the fourth electromagnetic signal generated by excitement of a fourth fluorophore of the multiple fluorophores;
a processor controlled analyzer to receive, from the multi-channel detector, a cumulative signal based on the first, second, third, and fourth electromagnetic signals and apply a decoding matrix to the cumulative signal to unambiguously detect the presence or absence of at least each of M analytes by associating, for each analyte, a first value in a first component of the cumulative signal and a second value in a second component of the cumulative signal, wherein each first value is an intensity or range of intensities and each second value is a wavelength or a range of wavelengths, and wherein the second values comprise the first, second, third, and fourth wavelengths, and the determination is made without immobilization, mass spectrometry or melting curve analysis; wherein for the positive integer M,
M=C*log2 (F+l),
F is a positive integer and is equal to the maximum cumulative intensity of the first component of the signal, for any second value, when all of the analytes are present, and
C=4, 5, or 6; and
wherein F+l is a positive integer and wherein F+l is a power of 2, wherein Mis greater than the number of the second values used to encode the analytes (C), the multi-channel detector comprises C channels, and M and C are positive integers.
wherein said method non-degenerately detects the presence or absence of the at least seven polynucleotide analytes in said single sample solution volume, in any combination of presence or absence, without requiring any step of immobilization of said polynucleotide analytes, physical separation of said polynucleotide analytes, or mass spectrometry.

36. Claim 19 of the ’797 Patent recites:

19. A system comprising:
a processor;
a display; and
a non-transitory computer readable medium storing instructions thereon that, when executed by the processor, cause the processor to:
obtain cumulative signal data from a digital PCR instrument with a sample volume, the digital PCR instrument comprising a

light source and a multichannel detector with C channels, and the sample volume comprising M fluorescently labeled polynucleotide analytes;

apply a decoding matrix to the cumulative signal data to unambiguously determine the presence or absence of at least each of the M fluorescently labeled polynucleotide analytes by associating, for each fluorescently labeled polynucleotide analyte, a first value in a first component of the cumulative signal data and a second value in a second component of the cumulative signal data, wherein each first value is an intensity or range of intensities and each second value is a wavelength or a range of wavelengths,

wherein for a positive integer M,

M=C*log2 (F+l),

F is the maximum cumulative intensity of the first component of the cumulative signal data, for any second value, when all of the analytes are present,

M, C, and F are each positive integers, and

wherein F+l is a positive integer and wherein F+l is a power of 2; and

plot, on the display, a representation of the first value and the second value;

wherein the cumulative signal data comprises:

a first electromagnetic signal at a first wavelength from the sample volume, the first electromagnetic signal generated by excitement of a first fluorescently labeled polynucleotide analyte of the six fluorescently labeled polynucleotide analytes;

a second electromagnetic signal at a second wavelength from the sample volume, the second electromagnetic signal generated by excitement of a second fluorescently labeled polynucleotide analyte of the six fluorescently labeled polynucleotide analytes;

a third electromagnetic signal at a third wavelength from the sample volume, the third electromagnetic signal generated by excitement of a third fluorescently labeled polynucleotide analyte of the six fluorescently labeled polynucleotide analytes;

a fourth electromagnetic signal at a fourth wavelength from the sample volume, the fourth electromagnetic signal generated by excitement of a fourth first fluorescently labeled polynucleotide analyte of the six fluorescently labeled polynucleotide analytes; and

wherein the second values comprise the first, second, third, and fourth wavelengths, and the determination is made without immobilization, mass spectrometry or melting curve analysis.

37. Claim 22 of the ’797 Patent recites:

22. A reaction mixture for unambiguously detecting a presence or absence of M non-immobilized polynucleotide analytes of a single sample, the reaction mixture comprising:

a plurality of non-immobilized hybridization probes,

wherein the reaction mixture is subjected to conditions such that the plurality of polynucleotide analytes are amplified;
wherein at least one of the plurality of oligonucleotide primers is used to amplify a region complementary of at least one of the plurality of hybridization probes;
wherein the plurality of hybridization probes comprise a first hybridization probe conjugated to a first fluorophore and a second fluorophore,
wherein the first fluorophore and the second fluorophore, when excited by a light source, emit light of different wavelengths, and
the plurality of hybridization probes further comprises a third hybridization probe and a fourth hybridization probe, wherein the third hybridization probe is conjugated to a third fluorophore and the fourth hybridization is conjugated to a fourth fluorophore,
wherein the third fluorophore and the fourth fluorophore, when excited by a light source, emit light of a same wavelength, and
wherein the third hybridization probe and the fourth hybridization probe are present at different concentrations in the reaction mixture;
optionally the plurality of hybridization probes further comprises a fifth hybridization probe and a sixth hybridization probe, wherein the fifth hybridization probe is conjugated to a fifth fluorophore and the sixth hybridization is conjugated to a sixth fluorophore;
wherein the fifth fluorophore and the sixth fluorophore, when excited by a light source, emit light of a same wavelength, and
wherein the fifth hybridization probe and the six hybridization probe are present at different concentrations in the reaction mixture,
wherein for the positive integer M,
M=C*log2 (F+l),
F is a positive integer and is equal to the maximum cumulative intensity of the first component of the signal, for any second value, when all of the analytes are present, and
C=4, 5, or 6; and
wherein F+l is a positive integer and wherein F+l is a power of 2, and
wherein M is greater than the number of the second values used to encode the analytes, the multi-channel detector comprises C channels, and M and C are positive integers.

38. Claim 30 of the ’797 Patent recites:

30. A kit for the unambiguously detecting a presence or absence of a plurality of polynucleotide analytes of a single sample comprising the components claim 24 and instructions for the use of the kit.

39. The ’797 Patent describes an innovative system for unambiguously identifying multiple targets in a single sample, even when multiple targets share the same

fluorescence color. Unlike traditional approaches that rely on a one-to-one correlation between fluorophores and targets, the '797 Patent discloses a multi-channel detection system that deciphers cumulative fluorescence signals across multiple wavelengths using a processor-controlled analyzer. By applying a decoding matrix, the system enables the identification of more targets than the number of available detection channels by leveraging cumulative signal intensities. This approach significantly improves the efficiency of multiplexed biochemical assays by maximizing detection capacity while minimizing spectral requirements. This innovation allows each cumulative signal to uniquely correspond to a specific target, despite multiple targets utilizing the same color channel, thereby overcoming the fundamental limitations of traditional fluorescence-based detection. The '797 Patent's system eliminates the need for immobilization, mass spectrometry, or melting curve analysis, allowing for real-time, high-throughput, and highly specific target detection—a breakthrough that others could not achieve.

40. The '797 Patent provides solutions to the long-existing problem of unambiguously identifying multiple targets without expanding the colors used. In some cases, it enables the detection of at least five or more targets using fewer than five colors (*e.g.*, five targets with four or fewer colors). Caltech's patented multiplexing solution represents a significant advancement over existing PCR technology, making highly efficient, commercially viable advanced PCR detection assays possible. This breakthrough has expanded the potential for real-world applications in critical areas, including the fight against diseases like cancer, where high-throughput, precise detection is essential.

THE INFRINGING PRODUCTS

A. Bio-Rad Launches its Infringing QX600 Droplet Digital PCR System

41. As of the time Caltech's inventors conceived of their HDPCR technology, Bio-Rad had also been trying to develop an advanced multiplexing technology of its own. In 2011, Bio-Rad researchers allegedly figured out a process of identifying more targets

than colors in a single sample and filed for patent protection of their process. *See* U.S. Provisional Patent Application No. 61/454,373, filed on March 18, 2011. Although Bio-Rad received patents on their approach to multiplexing, *see* U.S. Patent Nos. 9,222,128 and 9,921,154, Bio-Rad was unable to unambiguously detect multiple targets per color or unambiguously detect more than four targets with two colors. Since that time, despite many years trying to make its own inferior multiplexing technology work, this limitation rendered Bio-Rad's approach commercially unviable.

42. Unable to make its own approach commercially viable, Bio-Rad instead turned to Caltech's patented HDPCR technology and incorporated key aspects of it into its QX600 Droplet Digital PCR System (the "QX600 System").[5] By late 2022, Bio-Rad announced its new QX600 System, which enables multiplexed detection beyond the number of available colors, a core feature described in the application leading to the '797 Patent and protected by other patents in the parallel proceedings. *See supra* ¶¶ 17-18. Bio-Rad's own public statements confirm that the QX600 System is capable of unambiguously identifying 12 targets using only 6 fluorescence colors—precisely the type of innovation protected by the '797 Patent. For example, in an April 5, 2023 press release, Bio-Rad touted:

> Bio-Rad Laboratories, Inc. (NYSE: BIO and BIOb), a global leader in life science research and clinical diagnostic products, is accelerating measurable residual disease (MRD) research through more than a half dozen collaborations with institutions and companies leveraging its new QX600™ Droplet Digital™ PCR System. ***Launched late last year, it boasts six color detection capable of quantifying 12 targets per well***, a simple user workflow, and powerful data analysis. Maintaining Bio-Rad's best-in-class ddPCR™ technology, this platform utilizes the same droplet generation and processing protocols as the

[5] As used herein, the term "QX600 System" includes Bio-Rad's QX600 Droplet Digital PCR System and related systems, components and assays, such as Bio-Rad's QX600 Auto DG Droplet Digital System.

> QX200™ system, enabling thousands of current customers to easily adopt its advanced multiplexing capabilities.[6]

43. A true and correct copy of Bio-Rad's April 5, 2023 press release, which is available at https://www.bio-rad.com/en-us/life-science-research/news/bio-rads-qx600-droplet-digital-pcr-system-advancing-measurable-residual-disease-research?ID=Bio-Rad-s-QX600-Drop_1680646933 (last visited February 18, 2025), is attached to this Complaint as **Exhibit B**.

44. Beginning at least as of December 14, 2022, Bio-Rad's website offered the QX600 System for sale, announcing: "The QX600™ Droplet Digital™ PCR System enables advanced six-color multiplexing, allowing clear discrimination of multiple targets with assays that are cross-compatible with the QX200™ Droplet Digital PCR System. The QX600 System is designed for researchers who need to quantify multiple targets with high accuracy, reproducibility, and sensitivity." A true and correct copy of the product page for the QX600 system is available at https://www.bio-rad.com/en-us/product/qx600-droplet-digital-pcr-system?ID=b07d12ac-0585-fc4c-a586-3ddf20d5c4a0 (last visited February 18, 2025) and is attached to this Complaint as **Exhibit C**. The QX600 System product page continues in the "Description" section:

> The QX600 Droplet Reader offers users:
> - Sensitive multiplexing
> - Six-color detection capability (FAM, HEX, Cy5, Cy5.5, ROX, and ATTO 590)
> - Quantification of up to 12 targets in a single well
> - Absolute quantification with 0.1% or better sensitivity
> - Sensitive and precise gene expression multiplexing

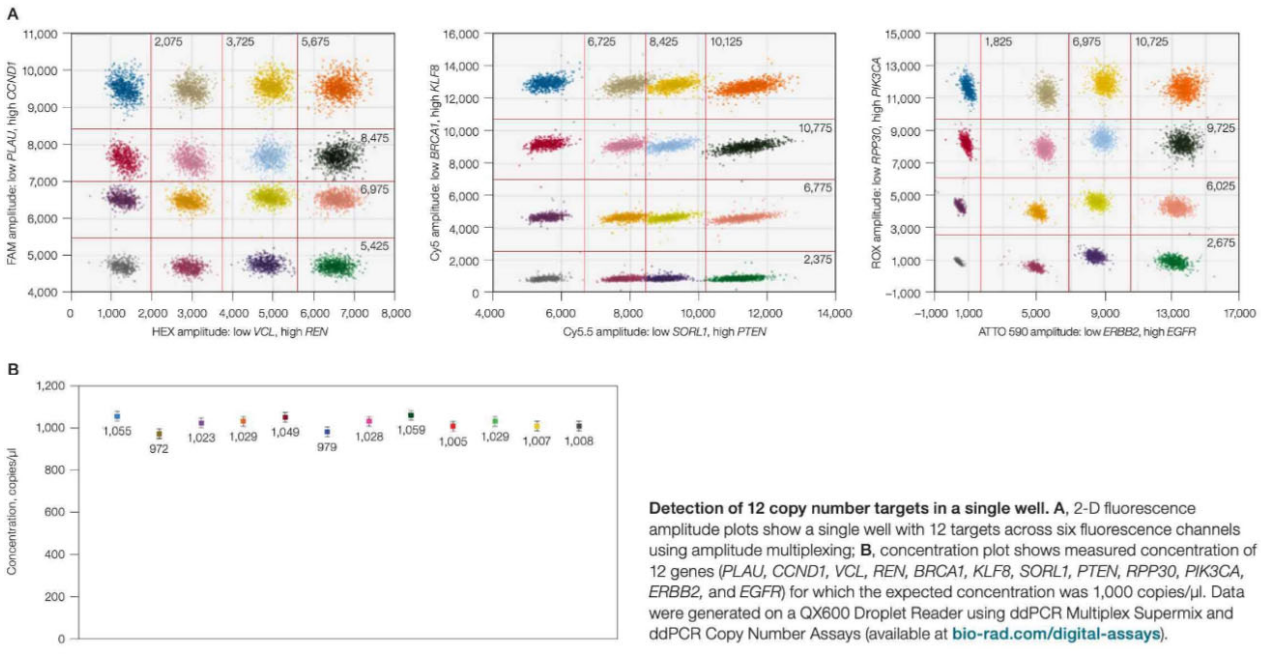
45. Bio-Rad also began distributing instructions on how to use the QX600 System and related assays to multiplex PCR processes to customers throughout the United

[6] Emphasis added.

States, including in this District, on its website, including the QX600 Droplet Reader and QX Manager Software Standard Edition User Guide (the "QX600 System User Guide"). A true and correct copy of the QX600 System User Guide is available at https://www.bio-rad.com/sites/default/files/2022-12/10000153877.pdf (last visited February 18, 2025) and attached to this Complaint as **Exhibit D**.

46. The QX600 System User Guide describes multiple assays for the QX600 that include an encoding scheme based on the cumulative signal from each fluorophore information that unambiguously identifies more targets than there are colors. For example, in Table 15, titled "Assay types," Bio-Rad describes assay type "Amplitude multiplex" as "Method to increase multiplexing up to twelve targets per well, with one or two targets detected per [color]." Ex. D at 77. Bio-Rad also describes a "Method assuming up to six probe colors … and up to six targets per [color]." *Id.*; *see also id.* at 79, Table 21 (describing "Fluorophore options" including "Amplitude multiplex, for 1 to 12 targets" and "Probe mix triplex, for 9 targets").

47. In Bio-Rad's "Bulletin 3557," a true and correct copy of which is attached to



this Complaint as **Exhibit E**, Bio-Rad provided data demonstrating their new and "unprecedented multiplexing capabilit[ies]," as reflected in the screenshot below labeled "Quantification of 12 Targets in a Single Well."

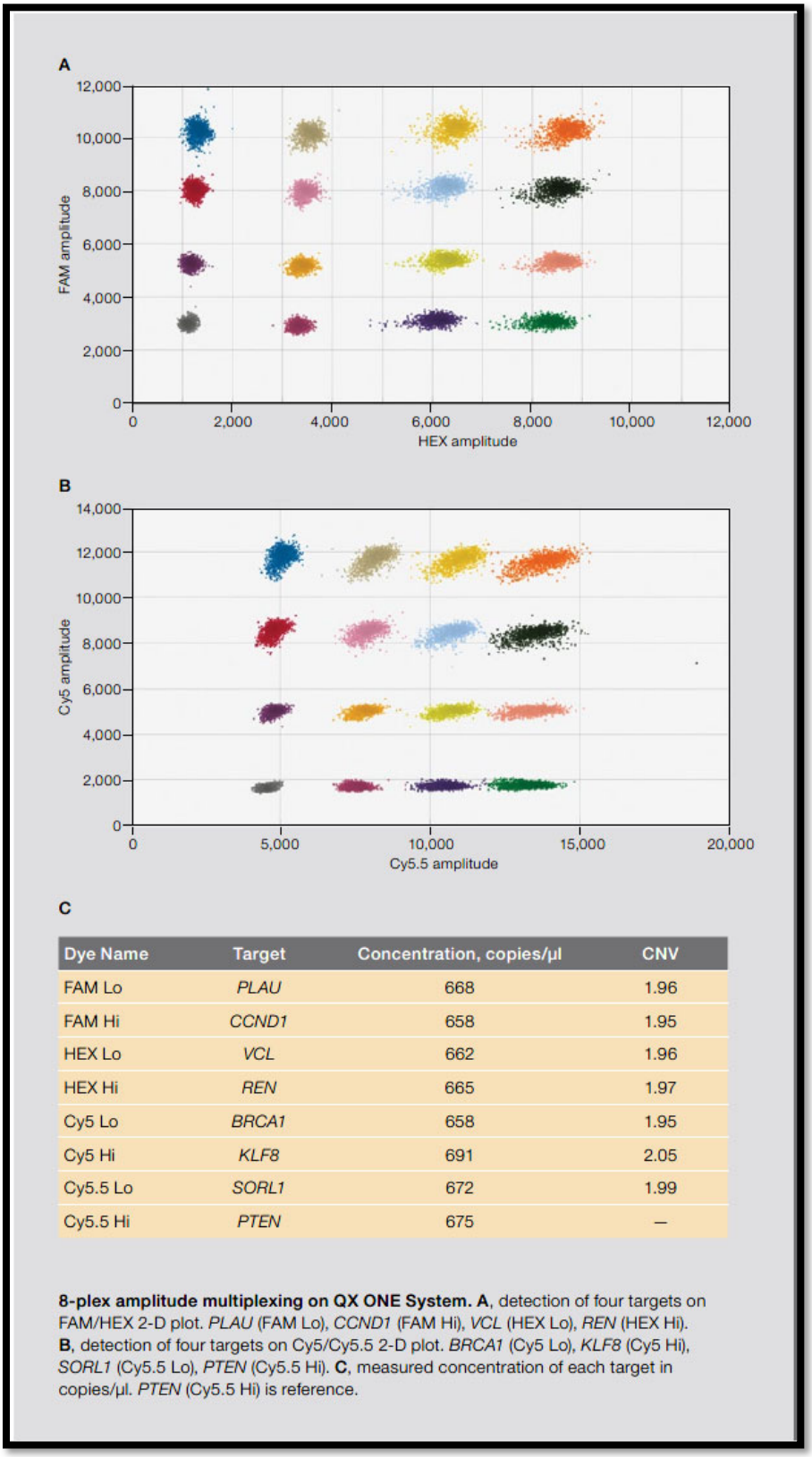
48. Bio-Rad continued to market and sell the QX600 System even after being put on notice of the issuance of the '797 Patent in December 2024. Despite knowledge of the '797 Patent and its relevance to Bio-Rad's QX600 System, Bio-Rad failed to obtain authorization and knowingly proceeded with its infringing activities. Accordingly, Bio-Rad has willfully infringed the '797 Patent since its issuance.

49. At least the QX600 System infringes one or more claims of each of the HDPCR Patents, including the '797 Patent. For example, the QX600 System's ability to unambiguously detect multiple targets per color using cumulative signals is only possible using ChromaCode's patented approach to multiplexing recited by one or more claims of each of the HDPCR Patents, including the '797 Patent. Additionally, Bio-Rad's QX600 System, when used with its associated reaction mixtures, meets the limitations of at least claim 22 of the '797 Patent. Bio-Rad supplies and promotes the use of reaction mixtures containing analyte-specific hybridization probes and multiple fluorophores as required by claim 22. Accordingly, Bio-Rad infringes claim 22 by making, selling, offering to sell, and inducing others to use such reaction mixtures in connection with the QX600 System. Thus, Bio-Rad's QX600 System infringes the HDPCR Patents, including the '797 Patent.

B. Bio-Rad'S QX ONE System Also Infringes the '797 Patent

50. Bio-Rad's QX ONE Droplet Digital PCR System and ddPCR Multiplex Supermix together also infringe one or more claims of the '797 Patent, as reflected in Bio-Rad's "Bulletin 6512," which is available at https://www.bio-rad.com/webroot/web/pdf/lsr/literature/Bulletin_6512.pdf (last visited February 13,

2025). A true and correct copy of Bulletin 6512 is attached as **Exhibit F** to this Complaint



C

Dye Name	Target	Concentration, copies/µl	CNV
FAM Lo	*PLAU*	668	1.96
FAM Hi	*CCND1*	658	1.95
HEX Lo	*VCL*	662	1.96
HEX Hi	*REN*	665	1.97
Cy5 Lo	*BRCA1*	658	1.95
Cy5 Hi	*KLF8*	691	2.05
Cy5.5 Lo	*SORL1*	672	1.99
Cy5.5 Hi	*PTEN*	675	—

8-plex amplitude multiplexing on QX ONE System. A, detection of four targets on FAM/HEX 2-D plot. *PLAU* (FAM Lo), *CCND1* (FAM Hi), *VCL* (HEX Lo), *REN* (HEX Hi). **B**, detection of four targets on Cy5/Cy5.5 2-D plot. *BRCA1* (Cy5 Lo), *KLF8* (Cy5 Hi), *SORL1* (Cy5.5 Lo), *PTEN* (Cy5.5 Hi). **C**, measured concentration of each target in copies/µl. *PTEN* (Cy5.5 Hi) is reference.

and a screenshot from that Bulletin is depicted above.

51. Bulletin 6512 states: "Bio-Rad's QX ONE Droplet Digital PCR System offers four color channels—FAM, HEX, Cy5, and Cy5.5—thereby providing additional multiplexing flexibility. By using Bio-Rad's ddPCR Multiplex Supermix on the QX ONE ddPCR System, as many as eight targets can be detected and measured in a single reaction." Ex. F at 9. Bulletin 6512 further states "Such advanced multiplexing is made possible using strategies such as amplitude multiplexing in conjunction with four color channels. Extract as much information as possible with high sensitivity, using as little sample as possible in a fast, cost-effective manner." Ex. F at 9. This further confirms Bio-Rad's unauthorized use of Caltech's patented technology to achieve multiplexed detection beyond the available number of fluorophore colors, a protected innovation under the '797 Patent.

52. Bio-Rad knew or should have known about Caltech's exclusive rights to practice one or more claims of the '797 Patent. Despite knowledge of Caltech's patent rights, Bio-Rad chose not only to infringe, but to induce others to do so as well. Indeed, as described above, Bio-Rad went so far as to provide step-by-step instructions on how to use Bio-Rad's devices to infringe one or more claims of the '797 Patent. This egregious misconduct has caused Plaintiff significant, irreparable harm that will continue unless and until Bio-Rad is enjoined from infringing the '797 Patent.

COUNT I

INFRINGEMENT OF THE '797 PATENT

53. Plaintiff repeats and realleges each and every allegation contained in the preceding paragraphs of this Complaint as if fully set forth herein.

54. Plaintiff is the owner of all right, title and interest to the '797 Patent.

55. Plaintiff's exclusive rights include the right to enforce the '797 Patent.

56. Bio-Rad makes, offers to sell and/or sells its infringing products in the United States, including in this District.

57. For example, Bio-Rad's QX600 System and QX ONE System (collectively, the "Accused Products") each practice each and every limitation, either literally or under the doctrine of equivalents, of at least claim 1 of the '797 Patent, in violation of 35 U.S.C. § 271(a). The Accused Products include a sample chamber configured to house a sample and analyte-specific reagent mixtures of analyte-specific hybridization probes and multiple fluorophores, a multi-channel detector that detects fluorescence signals at multiple wavelengths, with each fluorophore generating a cumulative fluorescence signal, a processor-controlled analyzer that receives cumulative fluorescence signals from the multi-channel detector and applies a mathematical model or equivalent decoding matrix to process the signals, enabling the detection of more analytes than the number of available fluorophore colors, as claimed in at least claim 1 of the '797 Patent. The Accused Products, therefore, directly infringe at least claim 1 of the '797 Patent.

58. Bio-Rad also indirectly infringes the '797 Patent. Bio-Rad has knowingly induced infringement of the '797 Patent by encouraging others to infringe at least claim 1 of the '797 Patent. For example, in Bio-Rad's QX600 System User Guide, Bio-Rad has induced, and will continue to induce, users to infringe the '797 Patent by expressly and intentionally instructing and encouraging users to perform each limitation of at least one claim of the '797 Patent in violation of 35 U.S.C. § 271(b). Bio-Rad is, therefore, liable for inducing infringement of the '797 Patent.

59. Bio-Rad's sale of Accused Products has contributed, and will continue to contribute, to the infringement of the '797 Patent in violation of 35 U.S.C. §271(c). For example, users of the QX600 System have infringed, and will continue to infringe, at least claim 1 of the '797 Patent by using the QX600 System in conjunction with assays designed to detect multiple targets per color using cumulative fluorescence signals, as described in the QX600 System User Guide. Bio-Rad is, therefore, liable for contributing to the infringement of the '797 Patent.

60. Upon information and belief, Bio-Rad had and continues to have knowledge

that multiple assays it provides for the Accused Products are especially made or especially adapted for a use that infringes the '797 Patent.

61. Upon information and belief, Bio-Rad had actual and constructive notice of the '797 Patent and knowingly or intentionally infringed the '797 Patent after acquiring that knowledge. Bio-Rad has no reasonable basis for asserting that the commercial manufacture, use, offer for sale, or sale of the Accused Products will not infringe, induce the infringement of, and/or contribute to the infringement of the '797 Patent.

62. Bio-Rad's infringement of the '797 Patent has been, and continues to be, willful, wanton, malicious, bad faith, deliberate, consciously wrong, flagrant, and egregious, entitling Plaintiff to an award of up to three times its actual damages pursuant to 35 U.S.C. § 284.

63. Bio-Rad's willful infringement of the '797 Patent further renders this an exceptional case under 35 U.S.C. § 285.

64. Plaintiff has been damaged by Bio-Rad's egregious and willful infringement of the '797 Patent.

65. Plaintiff will be irreparably harmed if Bio-Rad is not enjoined from infringing, and from actively inducing or contributing to the infringement of, the '797 Patent. Plaintiff does not have an adequate remedy at law, and, considering the balance of hardships between Plaintiff and Bio-Rad, a remedy in equity is warranted. Further, the public interest would not be disserved by the entry of a permanent injunction.

PRAYER FOR RELIEF

66. WHEREFORE, Plaintiff respectfully requests the following relief:

a. That the Court enter judgment that Bio-Rad directly infringed and infringes (literally and/or under the doctrine of equivalents), contributorily infringed and infringes, and induced and induces infringement of the '797 Patent in violation of 35 U.S.C. § 271;

b. That, prior to the expiration of the '797 Patent, the Court enjoin Bio-Rad from: (1) making, offering for sale and selling its infringing products and related multiplexing assays, and (2) infringing the '797 Patent;

c. That the Court award Plaintiff a reasonable royalty under 35 U.S.C. § 284 for Bio-Rad's infringement of the '797 Patent, beginning no earlier than the patent's issuance date and continuing through the expiration of the patent;

d. That the Court order an accounting to determine the full amount of reasonable royalties owed to Plaintiff under 35 U.S.C. § 284, including: (1) complete assessment of all infringing sales from the date of the '797 Patent's issuance through the date of judgment; (2) supplemental damages for any infringing sales not included in the initial damages award; and (3) ongoing royalties for post-judgment infringement if Bio-Rad continues to sell infringing products after the Court's final ruling.

e. Monetary damages to be awarded to Plaintiff as a result of Bio-Rad's infringing activities, including an accounting for infringing conduct not presented at trial and an award of additional damages for any such infringing activities;

f. That the Court find that Bio-Rad's infringement of the '797 Patent was egregious and willful and award Plaintiff three times their actual damages;

g. That the Court declare that this case is exceptional under 35 U.S.C. § 285 and award Plaintiff their attorneys' fees, costs, and expenses incurred in this action; and

h. That the Court award Plaintiff any further and additional relief as the Court may deem just, equitable, and proper.

JURY DEMAND

Plaintiff hereby demands a jury trial on all issues and claims so triable.

Dated: February 18, 2025

SHEPPARD, MULLIN, RICHTER & HAMPTON LLP

By */s/ Martin R. Bader*
MARTIN R. BADER

Attorneys for Plaintiff California Institute of Technology