

Bio-Rad Laboratories, Inc.

v.

California Institute of Technology

**Case No. IPR2024-01451
U.S. Patent No. 11,827,921 B2**

IN THE PATENT TRIAL & APPEAL BOARD

Claim Construction

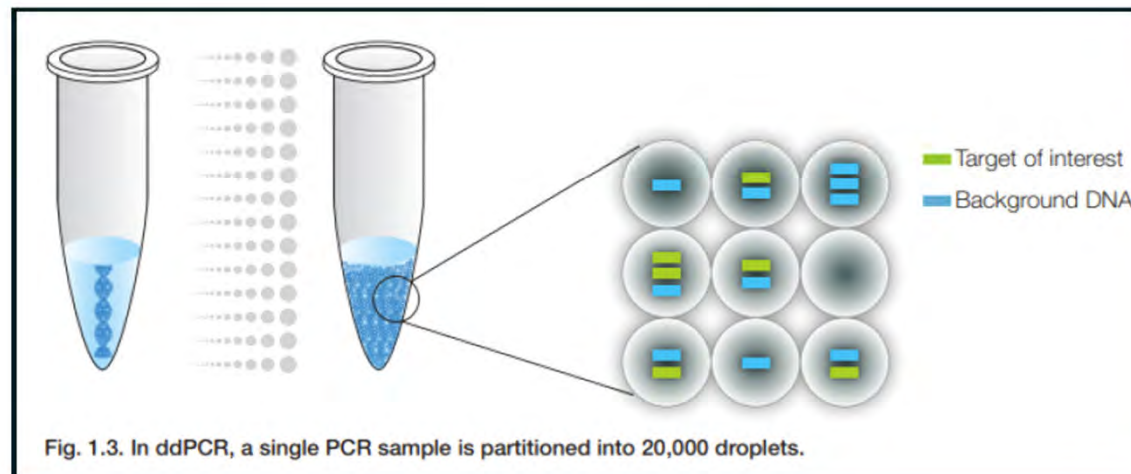
“single sample volume”

Claim Term	Bio-Rad’s Proposed Construction	Plaintiffs’ Proposed Construction
“single sample volume”	Plain and ordinary meaning	The “claimed ‘single sample volume’ <u>does not</u> include a volume that is subsequently partitioned into droplets, but, consistent with the express claim language, is where the reaction occurs (e.g., a droplet or well).” POR at 17.

PO's Original Infringement Contentions

“Single sample volume” may include a volume that is “subsequently” partitioned

For all of Bio-Rad's infringing ddPCR assays and kits, the detection is performed in a single sample volume that is subsequently partitioned into multiple droplets.



PO's Proposed Amended Infringement Contentions

PO belatedly sought to narrow claim scope

THE COURT: At page seven, Plaintiffs proposed removing all instances of "the detection is performed in a single-sample volume that is subsequently partitioned into multiple droplets" and replacing such instances with "the analytes are amplified in the single sample volume, such as in a droplet or well." That's not deleting and leaving the -- one -- you know, the second alternative or the first alternative in there. It's changing it.

EX1045 at 10:5-13

Patent Owner's Response

PO confirms that its proposed amendments sought to narrow claim scope

relies. This further reduces the weight (if any) that Petitioner or the Board should place on the extrinsic evidence from the preliminary infringement contentions. For example, the amendments clarify the claimed “single sample volume” does not include a volume that is subsequently partitioned into droplets, but, consistent with the express claim language, is where the reaction occurs (e.g., a droplet or well).

Still further, it is clear from PO’s preliminary infringement contentions regarding “wherein the analytes are amplified in the single sample volume” that the single sample volume PO accuses is where amplification occurs: e.g., in a droplet, not in a pre-partitioned volume. EX1012 at 27-29 (*citing* BIO_CC00007427 at 7450 (“droplets are run through a thermal cycler, which performs PCR amplification of the nucleic acid target in each individual droplet”)).

District Court Hearing On Motion To Amend Contentions

PO disavows narrowing construction of “single sample volume”

MR. SALEN: Your Honor, I don't know that the PTAB stated a claim construction, but we certainly don't have any issue with arguing the invalidity -- we don't take any point -- the Defendants argued in their IPR briefing and -- in the petition, and also the PTAB recognized that there are two different ways to read that claim. And we don't -- we're not trying to narrow a claim construction in the IPR based on our infringement contentions. What we're saying --

THE COURT: Yeah.

MR. SALEN: -- is that we don't have any evidence that one of those two alternatives actually occurs in the products. Those to us are different things. So we're not trying to narrow something -- we're not trying to narrow construction. We're adjusting the way we're reading that claim.

District Court Hearing On Motion To Amend Contentions

PO disavows narrowing construction in district court

THE COURT: Okay. All right. Let me just move on and -- make sure I get an answer to the question about the construction. So the construction that the PTAB assumed, you would be fine adopting that for purposes of anything in the case? **The two embodiments are within the scope.** So whether it's invalidity or something, I -- what we -- what I'm wondering is, is there an O2 Micro problem here where you're ultimately going to end up asking the jury to interpret it if you don't do something in advance?

MR. SALEN: I mean, **we think plain and ordinary meaning is the right** --

THE COURT: But that doesn't tell me anything. What does plain and ordinary meaning mean?

MR. SALEN: Yes, your Honor, is the answer. **Yes. We don't have a problem with the reading of the claim the way that the PTAB has done it.** We're not trying to undo something --

THE COURT: Okay.

MR. SALEN: -- that's been done in invalidity. We're just trying to clarify how we're reading the claim for the infringement based on the evidence that we've seen in the case.

District Court Hearing On Motion To Amend Contentions

PO waived any narrowing construction

THE COURT: ... Is that your plan? That you get this amendment, and then you go to the PTAB, and you say, "Oh, our narrower interpretation and our narrower infringement theory has been blessed, and we've been allowed to proceed, so we no longer have this broader interpretation." Is that the plan?

MR. SALEN: No, your Honor, our -- nothing has changed from how we're arguing in the IPR and nothing will change how we've been arguing in the IPR.

Supp Claim Construction Br. at 10

EX1045 at 56:14-21

PO's Supplemental Claim Construction Brief

PO disavows narrowing construction

litigation. To the contrary, PO agrees with Petitioner that the '921 Patent's claim term "single sample volume" should be given its plain and ordinary meaning, and accordingly that "single sample volume" should be broadly read as "*not* limited to an individual 'droplet,' but rather includes any type or nature of volume, such as a 'tube or bottle.'" Paper 10 at 3 (quoting Paper 9 at 1, 2); Paper 25, 4–7, 10. And

Supp Claim Construction Br. at 1

PO's Resp CC Br. at 5

PO's Supplemental Claim Construction Brief

PO disavows narrowing construction

“wherein” clause. Paper 25 at 1 (citing Paper 17 at 26). Indeed, PO agrees with Petitioner that the term “single sample volume” by itself can include “volumes that are subsequently partitioned into droplets.” Accordingly, there is no need for the Board to construe this term. *Nidec Motor Corp. v. Zhongshan Broad Ocean Motor*

Supp Claim Construction Br. at 1

PO's Resp CC Br. at 5

Claim 1 of the '921 Patent

Claim language does not limit “sample volume”

1. An assay that is capable of unambiguously detecting the presence or absence of each of at least M analytes, wherein each of said at least M analytes is encoded as a value of one component of a fluorescent signal, thereby generating a coding scheme, wherein said fluorescent signal comprises F wavelength components, and wherein said assay is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in a single sample volume, based on detection of said F wavelength components, when M is greater than F, wherein the assay does not require the use of mass spectrometry or immobilization of said at least M analytes, and wherein the analytes are amplified in the single sample volume.

'921 Patent Specification

Confirms that volume can be “tube or a bottle”

fication reaction 604. In the embodiment depicted in 601, the reagents for the detection of analytes and the primers are each contained in a single volume (e.g., tube or a bottle).

Reply at 10

EX1001 ('971 Patent) at 38:63-65

'921 Patent Specification

Confirms that separation need not be used

a solid phase. Of course, one of ordinary skill in the art would understand that the invention could be used with separation, beads, or a solid phase, if desired.

Reply at 10

EX1001 ('971 Patent) at 36:4-6

tive measurement on a single solution. No separation is necessary. The invention does not require the use of beads or

Supp Claim Construction Br. at 6

EX1001 ('971 Patent) at 36:2-3

absence, in a single sample volume without immobilization, separation, mass spectrometry, or melting curve analysis. In

Supp Claim Construction Br. at 6

EX1001 ('971 Patent) at 1:56-57

Dr. Batt

Confirms that separation need not be used



Petitioner's Expert:
Carl Batt PhD

14. The claims and specification do not prohibit analytes from being compartmentalized or separated for analysis. This can be done using a widely used technique called “droplet-based microfluidics,” which involves separating a single sample volume into uniform droplets ranging from picoliters to nanoliters, all of which are then suspended in an immiscible fluid and analyzed *en masse*. The '921 specification describes both separated and non-separated scenarios, stating “the invention *could* be used with separation ... if desired.” *Id.* at 36:4–6.

Supp Claim Construction Br. at 6

EX1043 Batt Decl. ¶ 14

“F”

Claim Term	Bio-Rad’s Proposed Construction	Plaintiffs’ Proposed Construction
“F”	Can be 1 or greater.	Must be 2 or greater.

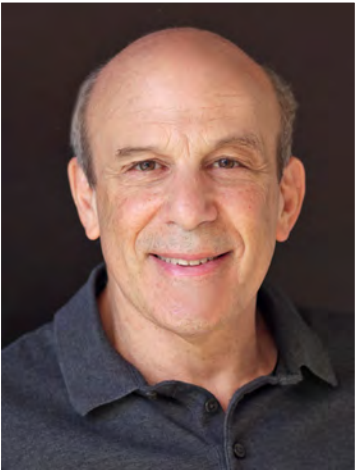
Claim 1 of the '921 Patent

Imposes no limits on minimum value of F

1. An assay that is capable of unambiguously detecting the presence or absence of each of at least M analytes, wherein each of said at least M analytes is encoded as a value of one component of a fluorescent signal, thereby generating a coding scheme, wherein said fluorescent signal comprises F wavelength components, and wherein said assay is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in a single sample volume, based on detection of said F wavelength components, when M is greater than F, wherein the assay does not require the use of mass spectrometry or immobilization of said at least M analytes, and wherein the analytes are amplified in the single sample volume.

Dr. Struhl

Confirms that “F could be 1”



PO's Expert:
Kevin Struhl PhD

- Q. Okay. Now, is there anything in the claim language that places any further constraints on the value of F?
- A. I don't think in the claim right here, there is no -- there is no number to put the M and F.
- Q. Okay. So you agree that in the claim, as the claim is written F can take on a value of 1?
- A. According to -- in this claim, F could be 1.
- Q. And then M could be 2?
- A. M could be 2, yes.

Reply at 8

EX1044, Struhl Depo. 85:9-22 (objections omitted)

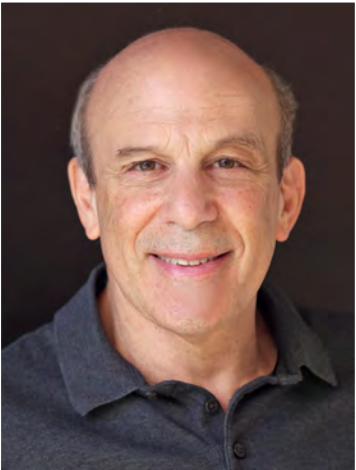
'921 Patent Specification

Teaches “single color coding scheme”

The coding scheme illustrated in Table 8 is non-degenerate by design. Although the coding scheme illustrated in Table 8 uses both intensity and color to encode each of the 16 analytes, each of the analytes could also be encoded by simply utilizing intensity. For example, given the 16 analytes provided in Table 8 (A-P), a single color coding scheme encoding all 16 analytes may assign the values 1, 2, 4, 8, 16, 32, 64, 128, 256, 512, 1024, 2048, 4096, 8192, and 16384 to analytes A, B, C, D, E, F, G, H, I, J, K, L, M, N, O, and P, respectively. Such a coding scheme is non-degenerate by design and the cumulative intensity result can be unambiguously decoded to indicate the presence or absence of analytes A-P. This coding scheme is capable of

Dr. Struhl

Agrees that specification's single-color approach can be used for '921 patent claims



PO's Expert:
Kevin Struhl PhD

Q. Okay. Now, is this idea, that's laid out in Column 22 and in Paragraph 43, is that an approach that can be used for the claimed invention of the '921 patent?

A. Yes.

Q. Do you know? It is?

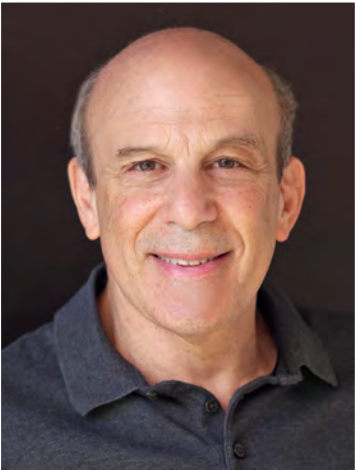
A. Yes. I don't see why not. Why you wouldn't -- you couldn't do that.

Reply at 8

EX1044, Struhl Depo. 68:15-23

Dr. Struhl

Agrees that specification's approach can be used with just two analytes



PO's Expert:
Kevin Struhl PhD

- Q. Okay. We'll come back to that, but I'm just asking about Column 22, Lines 18 to 37. You could do that with two analytes, right?
- A. You could put two different intensities in, yes.
- Q. You could put the first intensity, 1, and the second intensity, 2, right?
- A. Yeah, you could. You put the two intensities in there, but this has to be done in the context of a whole bunch of analytes floating around in there.

Reply at 8

EX1044, Struhl Depo. 70:16-71:1

Law

Excluding preferred embodiments is “rarely, if ever correct”



“A claim construction that excludes a preferred embodiment...is rarely, if ever correct and would require highly persuasive evidentiary support.”

Epos Techs. Ltd. v. Pegasus Techs. Ltd., 766 F.3d 1338, 1347 (Fed. Cir. 2014) (cleaned up)

Grounds 1 and 2: Invalidity In View Of Larson

PO's Infringement Allegations

Relies upon several coding tables

Analyte	FAM	HEX
N2	1	0
E	1	1
MHV	0	1

Petition at 11-12

EX1012 at 11

PO's Sur-Reply

Relies upon district court infringement contentions for non-obviousness

C. Petitioner's Arguments Regarding Objective Indicia of Nonobviousness Should Be Rejected

Petitioner argues there is no credible basis to conclude Bio-Rad's products infringe, and thus no evidence of commercial success or industry praise. Paper 20 at 24. This argument should be rejected because PO's infringement contentions provide this very basis and Petitioner has not refuted that they show infringement.

EX1012. Petitioner's attacks on PO's arguments for long-felt need are unpersuasive, because they rely on Petitioner's baseless attacks on Dr. Struhl's opinions. Paper 20 at 24-25. As explained herein, Dr. Struhl's opinions are reliable and should be credited. For these and the reasons set forth in PO's Response, objective indicia of nonobviousness support patentability.

Sur-reply at 23-24

Larson

Discloses allegedly infringing approach

Analyte	FAM	HEX
N2	1	0
E	1	1
MHV	0	1

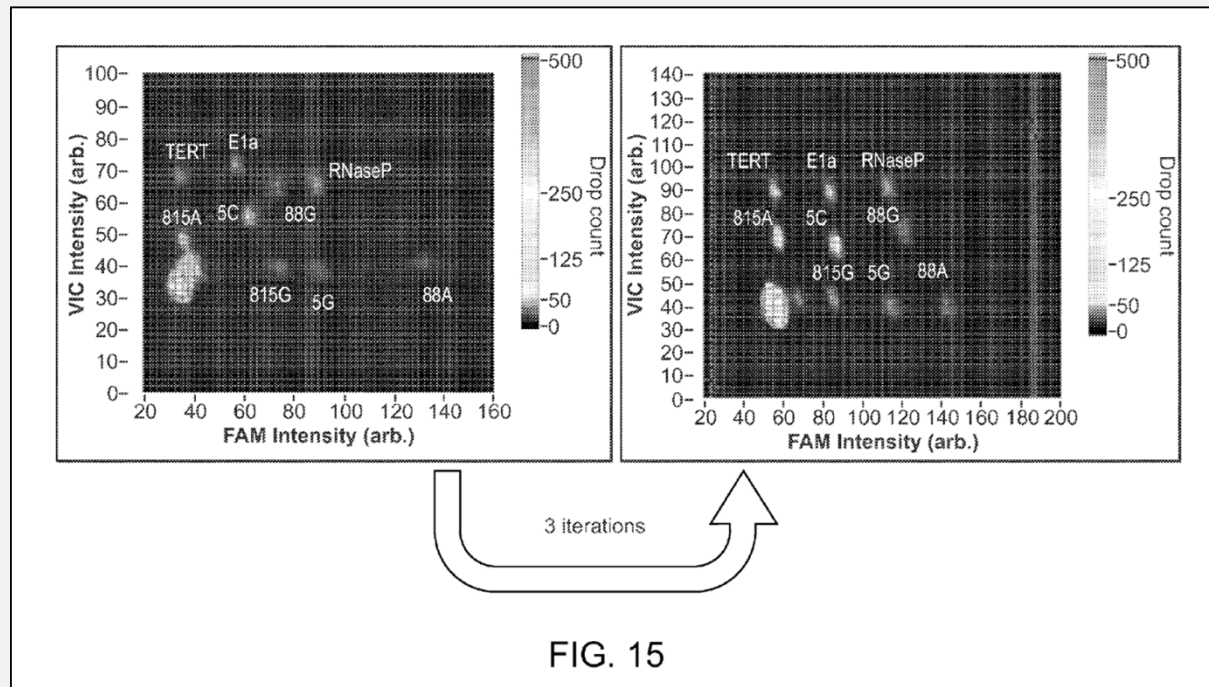
Petition at 11-12

EX1012 at 11

[0140] Another method of the invention involves performing higher-plex assays using multiple different probe colors (i.e. fluorophores), however unlike the strategy above where each target is identified by single type of probe with a unique color and intensity, instead in this method a single target may be identified by multiple probes that constitute a unique signature of both colors and intensities. For example, a single droplet might contain four different probes for measuring three different targets (say, T1, T2, and T3). Two probes might be of color A (say, A1, and A2), and two probes might be of color B (say, B1 and B2). T1 is measured by probe A1, T2 is measured by probe B1, but T3 is measured by both probes A2 and B2. Thus, when a droplet contains T1 only increased fluorescence appears in color A. When a droplet contains T2 only increased fluorescence appears in color B. However when a droplet contains T3, increased fluorescence appears in both colors A and B.

Larson

Teaches unambiguous detection of far more than just three targets



Petition at 45-46

EX1017 Larson Fig. 15 (Petition at 32)

Patent Owner's Response

Relies upon theory that “single sample volume” is limited to a droplet

1. Ground 1, Claims 1-10, 13-19: Anticipation by Larson.

Petitioner alleged Larson anticipates claims 1-10 and 13-19. Petitioner's arguments lack merit for at least the following reasons.

a. The challenged claims and Larson differ in kind, not just degree.

The '921 Patent and Larson take opposite approaches to extend the number of analytes that can be examined simultaneously. EX2018, ¶¶83-95.

While the '921 Patent is directed to detecting multiple analyte molecules in a single sample solution, Larson can only unambiguously distinguish analyte molecules from different sample volumes (i.e., droplets). This is because Larson is expressly “based on the singular nature of amplifications at terminal or limiting dilution that arises because most often only a single target allele is ever present in any one droplet even when multiple primers/probes targeting different alleles are present.” EX1017, ¶45; *id.*, ¶¶98-99; EX2018, ¶84; EX2017, 102:21-104:9. That is, in Larson, the samples are sufficiently diluted before PCR that for purposes of detecting analytes, most droplets before PCR either contain no analyte molecules or only a single analyte molecule. EX2018, ¶84.

Patent Owner's Sur-Reply

Relies upon theory that “single sample volume” is limited to a droplet

B. Grounds 1, 2, and 4 Fail

1. Larson does not anticipate the challenged claims

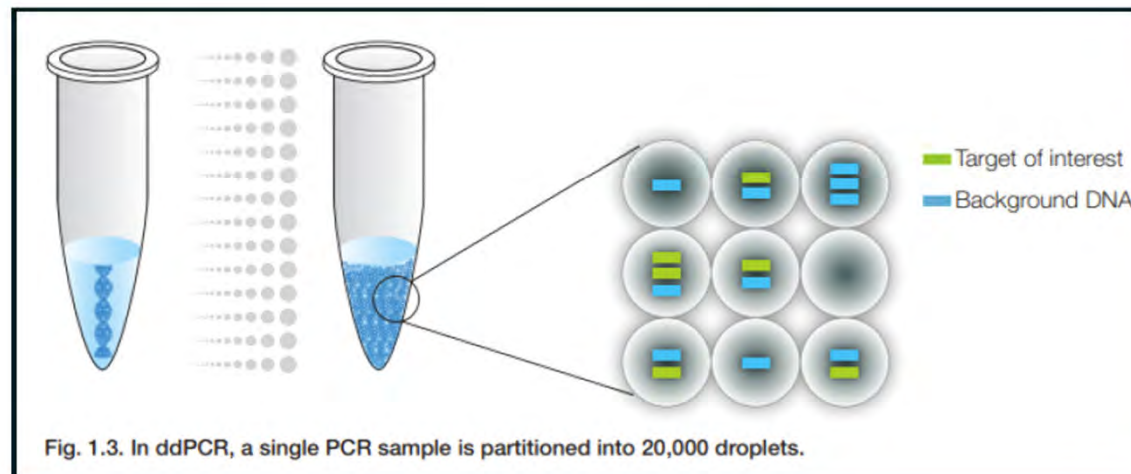
PO's Response explains that Larson does not anticipate (Ground 1) because FIG. 9C and ¶121 do not disclose using two or more colors to detect three or more analytes in the same single sample volume in which amplification occurs, which the '921 Patent claims require and on which Petitioner's arguments focus. Paper 17 at 12-13, 27-36; Paper 20 at 10; EX1043, ¶¶13, 20-21, 27; EX2025, 23:10-25. Petitioner accuses PO of taking “an overly-narrow reading of Larson that disregards the reference's explicit disclosures.” Paper 20 at 23. But PO has done no such thing. Larson lacks such teachings, and even Dr. Batt ultimately conceded that Larson's ¶121 “doesn't account for” multiple-occupancy microdroplets containing targets 1+2, 1+3, 2+3, or 1+2+3” and that FIG. 9C is a *single-occupancy* embodiment. EX2017, 108:4-109:4, 113:6-15, 114:13-115:23, 210:12-

Sur-Reply at 19

PO's Original Infringement Contentions

Alleges that “single sample volume” may be a volume that is “subsequently” partitioned

For all of Bio-Rad's infringing ddPCR assays and kits, the detection is performed in a single sample volume that is subsequently partitioned into multiple droplets.



The Petition

Grounds 1 and 2 do not rely upon a droplet as the “single sample volume”

is two. EX1002 ¶ 130. Larson discloses detecting whether these three targets are present in a “sample volume,” specifically, a portion of a sample solution that is subsequently partitioned into droplets. *See id.* ¶¶ 131-132; EX1017 ¶¶ 11, 13-17, 21-23, 52-68, 154, 164, Figs. 1-3.

Petition at 29

Claim 1 of the '921 Patent

Requires that analytes be “amplified in the single sample volume”

1. An assay that is capable of unambiguously detecting the presence or absence of each of at least M analytes, wherein each of said at least M analytes is encoded as a value of one component of a fluorescent signal, thereby generating a coding scheme, wherein said fluorescent signal comprises F wavelength components, and wherein said assay is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in a single sample volume, based on detection of said F wavelength components, when M is greater than F, wherein the assay does not require the use of mass spectrometry or immobilization of said at least M analytes, and wherein the analytes are amplified in the single sample volume.

Larson

Explains that the droplets are collected and amplified en masse

[0079] Once final droplets have been produced by any of the droplet forming embodiments above, or by any other embodiments, the droplets are thermal cycled, resulting in amplification of the target nucleic acid in each droplet. In certain embodiments, the droplets are collected off-chip as an emulsion in a PCR thermal cycling tube and then thermally cycled in a conventional thermal cycler. Temperature profiles for thermal cycling can be adjusted and optimized as with any conventional DNA amplification by PCR.

Reply at 23

EX1017 Larson ¶ 79

Larson

Explains that the droplets are collected and amplified en masse

[0193] Approximately 25 uL of the PCR reaction mixture was collected as an emulsion from the droplet generation chip and thermally cycled in a DNA Engine (Bio-Rad, Hercules, Calif.). The reaction mixture contained 1× TaqMan® universal PCR master mix (Applied Biosystems, Carlsbad, Calif.), 0.2 mM dNTP (Takara Bio, Madison, Wis.), and various amounts of primer pairs and probes as described in the results. 1× assay concentration is defined as 0.2 μM probes with 0.9 μM primers. In all cases, when varied from the 1× concentration, the primers and probes were varied by the same amount. The cycler program included a 10 min hot start at 95° C., and 45 cycles of 15 s at 95° C. and 60 s at 60° C.

Reply at 23

EX1017 Larson ¶ 193

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Petitioner's Expert

Confirms that droplets are collected for amplification



Petitioner's Expert:
Carl Batt PhD

34. Larson also teaches amplification within a “single sample volume,” even under Patent Owner’s construction of the term. This can be seen in Larson’s description of droplet formation. *See* EX1017 at ¶¶ 78–79, 193. Larson describes taking a sample and separating the sample into droplets. *Id.* Following droplet formation, Larson teaches “the droplets are collected off-chip as an emulsion in a *PCR thermal cycling tube*.” *Id.* at ¶ 79. This tube of droplets is thermocycled in a thermal cycler, where DNA amplification occurs. *Id.* By amplifying the collected droplets, Larson ultimately describes a method where amplification occurs within a “single sample volume,” *i.e.*, the original sample volume in collected, compartmentalized form.

'921 Patent Claims 4 and 13

Claims 3 and 4 pertains to avoiding degeneracy

4. The assay of claim 2, wherein said coding scheme is made **nondegenerate** by enumerating every legitimate result that can be obtained from said coding scheme, identifying each legitimate result that is degenerate, and eliminating at least one potential analyte code from said coding scheme to eliminate degeneracy.

13. The assay of claim 1, wherein said coding scheme is innately **nondegenerate** or non-degenerate by construction.

Larson

Discloses adjusting the intensity values from color labels to avoid potentially ambiguous results

[0035] FIG. 15 depicts a 9-plex dPCR assay for spinal muscular atrophy with only two fluorophores, showing the process of optimizing droplet intensities. FIGS. 15a and 15b show 2-D histograms of droplet fluorescence intensity, shown as heat maps with hotter colors representing higher droplet counts, for the 9-plex assay against the synthetic model chromosome ((a)=Before optimization; (b)=after optimization).

[0146] As another method of the invention, histogram-based data presentation and analysis is incorporated into the invention for identifying and characterizing statistically similar populations of droplets that arise from one probe signature (color and intensity), and for discriminating one population of droplets from the others. FIG. 14a shows a 2-dimensional histogram of droplet fluorescence intensities as a contoured heat map, with hotter colors representing higher occurrences. Standard techniques were used to compensate for spectral overlap of the FAM and VIC signals. Samples were run at 0.006 occupancy per target. Six populations were clearly evident, five for the assay and one for PCR(-) droplets. As one method of the invention, the populations were assigned by selective exclusion of assay components. For example, excluding the SMN2 primers and probe eliminated the population at the bottom right in the histogram, but otherwise the distribution remained unchanged. Assignments are labeled in FIG. 14a. As we have found to be generally true for this method of multiplexing, the assay worked immediately with well resolved or at least distinguishable populations for each target. As another method of the invention, the relative positions of the different populations in the histogram were then adjusted into a regularly spaced rectangular array by tuning the probe concentration as described in the previous section. Usually no more than two iterations are required for optimization.

Petition at 34; Reply at 14

EX1017 (Larson US 2011/0250597) ¶ 35

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

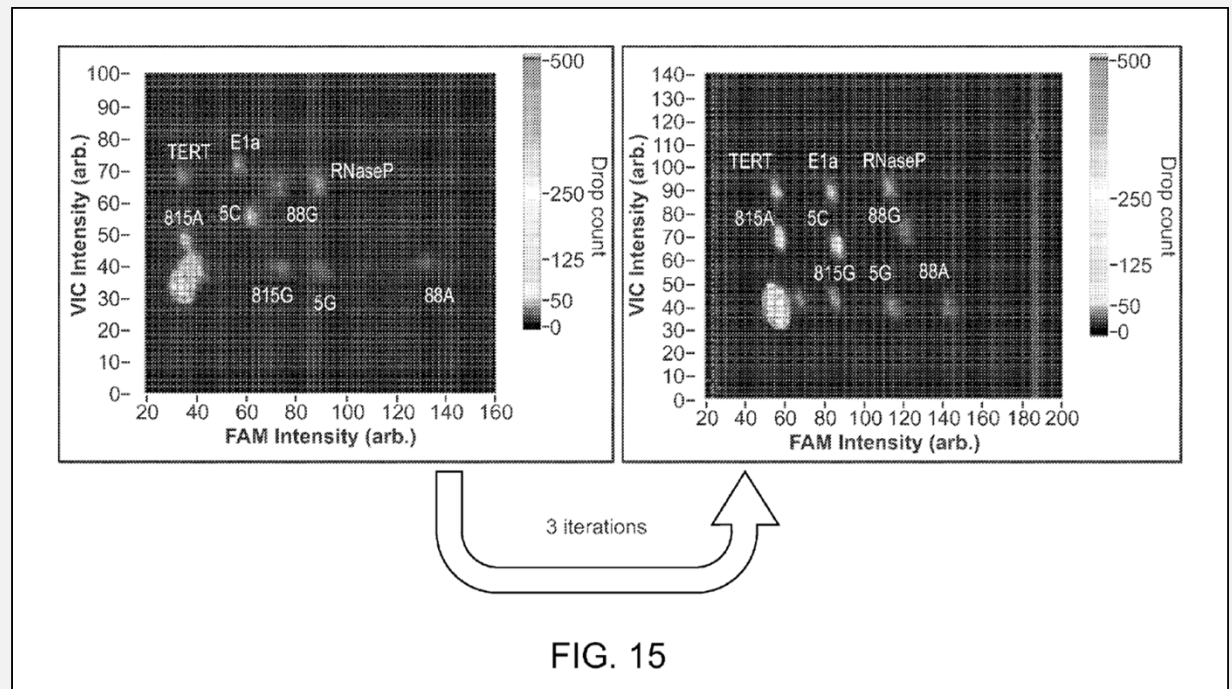
Petition at 34; Reply at 14

EX1017 Larson ¶ 146

Larson

Discloses adjusting the intensity values from color labels to avoid potentially ambiguous results

[0150] A 9-plex assay for certain SMA related targets was also demonstrated with just two colors (probes containing FAM and VIC fluorophores). Aside from the optimized primer and probe concentrations, assay conditions and experimental procedures were identical to the 5-plex assay above. FIG. 15a shows the various droplet populations in 2-D histograms before optimization of probe concentrations. The identity of the different targets is shown on the figure itself. As one method of the invention, the identification of the different populations was made as before, by selective exclusion and/or addition of one or more assays. Most of the populations were already well resolved, with the exception of the probe for the c.815A genotype that was in close proximity with the cluster corresponding to empty droplets. After three iterations of optimization of probe concentrations, all of the target populations were well resolved from each other, and well resolved from the empty droplets (FIG. 15b). Three methods of the invention were highlighted in this demonstration: (1) nine DNA targets were uniquely identified in a two-dimensional histogram, far beyond the capabilities of conventional qPCR; (2) target DNA molecules were distinguished on the basis of some combination of both color and intensity arising from one or multiple probes against the same target; and (3) the relative positions of the target molecules within the histogram were adjusted by varying the probe concentrations to optimize the pattern of colors and intensities for increased resolution amongst the various droplet populations.



Petition at 45-46

EX1017 Larson Fig. 15

Petition at 45-46

EX1017 Larson ¶ 150

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Petitioner's Expert

Confirms that optimization is functionally no different than eliminating analyte code



Petitioner's Expert:
Carl Batt PhD

292. A POSA would have understood that this optimization process is functionally no different than eliminating an analyte code that is degenerate to remove the degeneracy. They would have understood that in the process of optimization, the optimized analyte codes are necessarily eliminated, and subsequently replaced, to eliminate degeneracy and detect the signals representing the targets unambiguously.

Petition at 46

EX1002 Batt Decl. ¶ 292

Larson

Discloses allegedly infringing approach

Analyte	FAM	HEX
N2	1	0
E	1	1
MHV	0	1

Petition at 11-12

EX1012 at 11

[0140] Another method of the invention involves performing higher-plex assays using multiple different probe colors (i.e. fluorophores), however unlike the strategy above where each target is identified by single type of probe with a unique color and intensity, instead in this method a single target may be identified by multiple probes that constitute a unique signature of both colors and intensities. For example, a single droplet might contain four different probes for measuring three different targets (say, T1, T2, and T3). Two probes might be of color A (say, A1, and A2), and two probes might be of color B (say, B1 and B2). T1 is measured by probe A1, T2 is measured by probe B1, but T3 is measured by both probes A2 and B2. Thus, when a droplet contains T1 only increased fluorescence appears in color A. When a droplet contains T2 only increased fluorescence appears in color B. However when a droplet contains T3, increased fluorescence appears in both colors A and B.

Petition at 25; Reply at 14

EX1017 Larson ¶ 140

Ground 3: Obviousness In View Of Larson Alone

Claim 1 of the '921 Patent

Claims detection of more analytes than detection colors

1. An assay that is capable of unambiguously detecting the presence or absence of each of at least M analytes, wherein each of said at least M analytes is encoded as a value of one component of a fluorescent signal, thereby generating a coding scheme, wherein said fluorescent signal comprises F wavelength components, and wherein said assay is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in a single sample volume, based on detection of said F wavelength components, when M is greater than F, wherein the assay does not require the use of mass spectrometry or immobilization of said at least M analytes, and wherein the analytes are amplified in the single sample volume.

Patent Owner's Response

Inventiveness is allegedly using fewer fluorophores than targets

fluorophores.” EX1001, 12:39-45; *see also id.*, 8:58-62. To overcome this limitation, the inventors developed a technique to non-degenerately identify analyte molecules in a sample, using *fewer* fluorophores than the number of analyte molecules (in contrast to prior multi-channel techniques). EX1001, 8:63-

Petition at 11

Paper 17 at 8

Dr. Batt's Declaration

States UNDISPUTED competencies of a POSA known as early as 2002

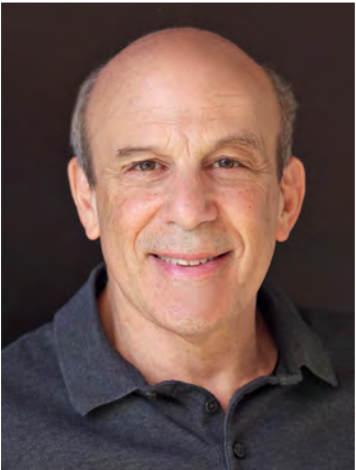
69. A POSA would have a strong understanding and/or experience with the design and application of various assays to detect polynucleotide sequences.

Specifically, this includes the following techniques:

- Multiplex PCR and its application in detecting and measuring polynucleotide analytes. *See, e.g., EX1017 ¶ 5* ("the ability to perform and ...
- The design and use of fluorescent-labeled oligonucleotide probes, including use of quenchers, and use in conjunction with multiplex PCR. ...
- The quantitative detection and measurement of polynucleotide analytes using fluorescent-labeled oligonucleotide probes in multiplex applications. ...
- Techniques for labeling probes and detecting multiple targets using fewer differently colored fluorophores than the number of polynucleotide targets. ...
- Quantitative PCR (qPCR) and digital PCR (dPCR) and their application in detecting and measuring polynucleotide analytes, including in multiplex applications. *See, e.g., id. ¶ 6* ("The principle advantage of digital

Dr. Struhl

Confirms competencies of a POSA known as early as 2002



PO's Expert:
Kevin Struhl PhD

Q. Okay. So let me ask this question: At what point in time would you have said that you would have understood the competencies that are part of **Petitioner's definition of the level of ordinary skill in the art?**

A. I would say that I would be a Person of Ordinary Skill in the Art in like **2002, 2003.**

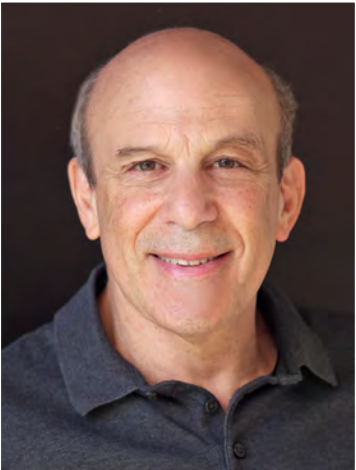
Q. Okay. That's when you would have understood the competencies that are listed in Petitioner's --

A. Yeah. **We were doing it.**

Reply at 11

EX1044, Struhl Depo. 38:5-15

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Would have understood competencies of POSA as of priority date

Q. And then recall, we went through several bullet points, the four bullet points --

A. Yes.

Q. -- that Mr. Walter showed you about the understanding of a person of the ordinary skill in the art?

A. Yes.

Q. Did you have those four bullet points, would you have understood those concepts as of February 3, 2012?

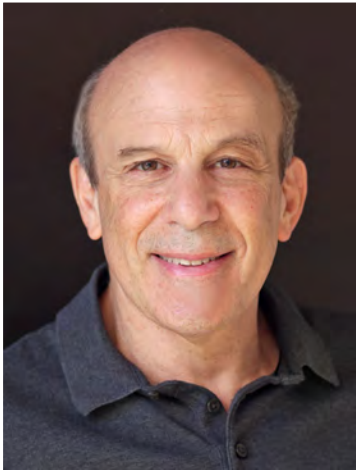
A. Yeah, because that's what I was judging it upon, and I would have judged it on that.

Reply at 11

EX1044, Struhl Depo. 194:13-24

Dr. Struhl

Confirms multiplex PCR being done as early as 2002

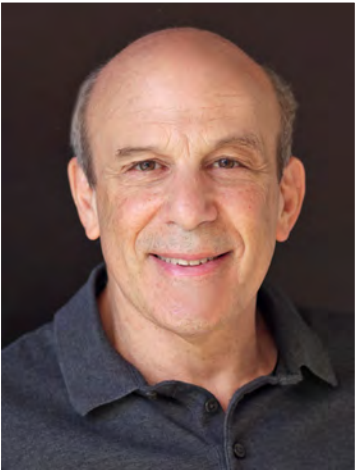


PO's Expert:
Kevin Struhl PhD

- Q. ... Okay. So the techniques that are listed in Petitioner's definition for the level of ordinary skill in the art, those are things that you and your coworkers were doing in the lab in the 2002, 2003 time frame?
- A. Yeah. It varies to exactly which things were done when. I couldn't give you an exact number, but we were about -- we were about as early as it gets in doing PCR reactions for various purposes.
- Q. Okay. Let's talk about some of these things. One of the competencies that's listed in the first bullet point, Petitioner's definition is multiplex PCR and its application detecting and measuring polynucleotide analytes. When did you first start doing multiplex PCR?
- A. Our first published paper that used it, I believe, was 2002. I think the 2002 paper has it in there. 2003 paper definitely has it in there.
- Q. Okay. And how many targets were you multiplexing in the same reaction?
- A. I think either -- a few. I think it was like three or five. Some number like that.

Dr. Struhl

Confirms multiplex PCR is “not magical”



PO's Expert:
Kevin Struhl PhD

Q. Okay. And in 2000 -- so you said you were doing this in 2002, 2003?

A. There are two papers. There are three papers I can think off the top of my head that did this; 2002, 2003, 2005. Paul Mason in my lab is the guy who was doing all of it.

Q. And when you went to 2000 -- when you were in 2005, how many targets were you doing?

A. Well, for multiplex PCR, we were typically doing, I'd say, probably -- five was probably what we usually routinely did. Maybe sometimes there might have been fewer but --

Reply at 12

EX1044, Struhl Depo. 40:8-19

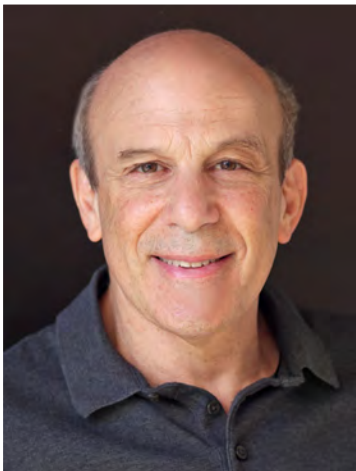
Q. Okay. Now, you said if -- you had done five. If you wanted to do seven, you would have done seven. Why are you confident in 2005, you would have done five or seven?

A. It's not magical.

Reply at 12

EX1044, Struhl Depo. 42:5-9

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Larson and Saxonov modulate intensities to distinguish targets

- Q. Okay. And do you know when that idea of using different intensities was first suggested?
- A. Actually, I don't. I certainly both -- certainly the -- certainly all the relevant art here, like Larson and Saxonov of the patent all do that, okay. That's when I first heard about it. I didn't -- I hadn't heard about it until then.

Reply at 12

EX1044, Struhl Depo. 51:15-21

- Q. Now, when did people start to use multiple colors or using multiple fluorophores to modulate the intensity; do you know when?
- A. All I know is what I've read in these. I did not go, and do a history of this. So it it's clear that the '921 patent and the Saxonov and Larson reference they do that at some level, and that's where I first heard about it. I don't know the history of that. I don't know if there is a paper before that saying you could do this, the other thing.

Reply at 12

EX1044, Struhl Depo. 52:9-18

Patent Owner's Response

Contends Larson is limited to single occupancy droplets

- a. **Larson Teaches Eliminating the Very “Multiple Occupancy” Situations to Which Petitioner Points.**

Petitioner's Ground 3 alleges Larson teaches “there may be multiple targets in a single individual droplet and that its methods accommodate such ‘multiple occupancy’ situations,” stating “Larson’s assays use conditions and parameters that do precisely this.” Petition, 52, *citing* EX1017, ¶¶129, 141; EX1002, ¶321.

However, **Larson’s actual disclosure about accommodating “multiple occupancy” situations” is *incompatible* with an assay that is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in the single sample volume in which the analytes are amplified.** EX2018, ¶99; *see* EX1001, 67:16-20, 68:31-34.

Larson

Teaches “limiting dilution” where droplets “frequently” have multiple molecules

[0006] Digital PCR (dPCR) is an alternative quantitation method in which dilute samples are divided into many separate reactions. See for example, Brown et al. (U.S. Pat. Nos. 6,143,496 and 6,391,559) and Vogelstein et al. (U.S. Pat. Nos. 6,440,706, 6,753,147, and 7,824,889), the content of each of which is incorporated by reference herein in its entirety. The distribution from background of target DNA molecules among the reactions follows Poisson statistics, and at so called “terminal dilution” the vast majority of reactions contain either one or zero target DNA molecules for practical intents and purposes. In another case, at so called “limiting dilution” some reactions contain zero DNA molecules, some reactions contain one molecule, and frequently some other reactions contain multiple molecules, following the Poisson distribution. It is understood that terminal dilution and limiting dilution are useful concepts for describing DNA loading in reaction vessels, but they have no formal mathematical definition, nor are they necessarily mutually exclusive. Ideally, at terminal dilution, the number of PCR positive reactions (PCR(+)) equals the number of template molecules originally present. At limiting dilution, Poisson statistics are used to uncover the underlying amount of DNA. The principle advantage of digital compared to qPCR is that it avoids any need to interpret the time dependence of fluorescence intensity—an analog signal—along with the main underlying uncertainty of non-exponential amplification during early cycles.

Larson

Teaches that the invention is compatible with limiting dilution

[0052] Methods of the invention involve forming sample droplets where some droplets contain zero target nucleic acid molecules, some droplets contain one target nucleic acid molecule, and some droplets may or may not contain multiple nucleic acid molecules (corresponding to limiting or terminal dilution, respectively, as defined above). In the preferred embodiment, the distribution of molecules within droplets obeys the Poisson distribution. However, methods for non-Poisson loading of droplets are known to those familiar with the art, and include but are not limited to active sorting of droplets, such as by laser-induced fluorescence, or by passive one-to-one loading. The description that follows assumes Poisson loading of droplets, but such description is not intended to exclude non-Poisson loading, as the invention is compatible with all distributions of DNA loading that conform to limiting or terminal dilution.

Larson

Teaches that his invention accommodates “multiple occupancy”

[0141] Generally, without wishing to be constrained by theory, the above three methods for higher-plex dPCR are simplest to implement under conditions of terminal dilution, that is when the probability of multiple different target molecules co-occupying the same droplet is very low compared to the probability of any single target occupying a droplet. With multiple occupancy arises the complexity of simultaneous assays competing within the same reaction droplet, and also complexity of assigning the resulting fluorescence intensity that involves a combination of fluorescence from two different reaction products that may or may not be equal to the sum of the two fluorescence intensities of the individual reaction products. However, methods of the invention can accommodate these complications arising from multiple occupancy.

Petition at 55

EX1017 Larson ¶ 141

Larson

Teaches detecting multiple targets in a single droplet

[0108] An example demonstration of droplet-based amplification and detection of multiple target sequences in a single droplet is shown here. Multiple copies of 5 sets of primers (primers for TERT, RNaseP, E1a, SMN1 and SMN2) were encapsulated in a single droplet at various concentrations along with the template DNA and the PCR master mix. Probes that specifically bind to TERT, RNaseP, E1a, SMN1 or SMN2 were also encapsulated in the droplets containing the primers. Probes for TERT, RNaseP and E1a were labeled with the VIC dye and probes for SMN1 and SMN2 were labeled with the FAM dye. The sequences for TERT, RNaseP, E1a, SMN1 and SMN2 were amplified by PCR. The PCR was conducted with a standard thermal cycling setting. For example:

[0109] 95° C. for 10 min

[0110] 31 cycles

[0111] 92° C. for 15 s

[0112] 60° C. for 60 s

[0113] At the end of the PCR, the fluorescence emission from each droplet was determined and plotted on a scattered plot based on its wavelength and intensity. Six clusters, each representing droplets having the corresponding fluorescence wavelength and intensity were shown. The TERT, RNaseP and E1a clusters showed the fluorescence of the VIC dye at three distinct intensities and SMN1 and SMN1 clusters showed the fluorescence of the FAM dye at two distinct intensities (FIG. 5). The number of droplets, each having one or more sequences selected from TERT, RNaseP, E1a, SMN1 and SMN2, can be determined from the scattered plot.

Reply at 16

EX1017 Larson ¶ 108-112

Reply at 16

EX1017 Larson ¶ 113

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Larson

Teaches droplets with multiple targets

[0129] Another advantage of the dPCR approach for multiplexed reactions is that the different reactions do not compete with each other for reagents as they would in a bulk qPCR assay. However, the possibility for unintended cross-reactivity remains. A multiplexed assay can require a more dilute sample. For instance, at 10% occupancy a duplex reaction would have double occupancy 1% of the time. Hence 1 in 10 PCR+ droplets would be doubles, resulting in a final intensity at least as high and possibly higher than the brighter of the two probes. For a simple duplex system the contribution from each probe could be recovered. In this example the total number of PCR+ droplets for probe 1 would be $(\text{Probe 1}) + (\text{Probe 1} + \text{Probe 2})$. Higher degrees of multiplexing would require greater dilution. For example, for a 4-plex at 1% occupancy the probability of one probe overlapping any of the other 3 is ~3%, and that error may be too high for some applications. The need for large dilutions strongly favors the large number of dPCR reactions.

Petition at 52

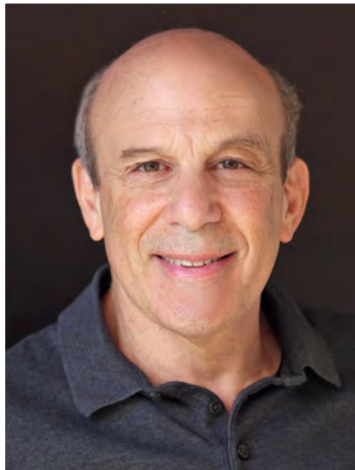
EX1017 Larson ¶ 129

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Struhl

Confirms that Larson teaches how to analyze droplets with multiple targets



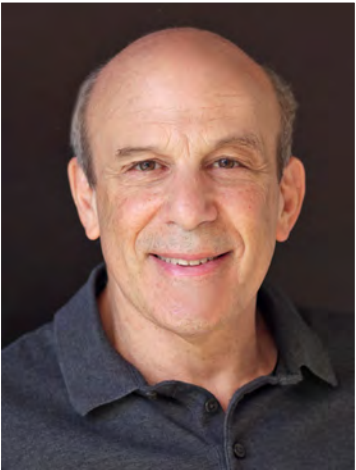
PO's Expert:
Kevin Struhl PhD

61. According to Larson's apparent definition of "occupancy," the term "10% occupancy" refers to a dilution at which 90% of droplets are empty, 10% of droplets contain analyte, and 10% of the droplets that are not empty (that is, 1% of all the droplets) contain exactly two analytes. *See id.*, ¶129. In this situation, in which the original sample must be very highly diluted, Larson teaches that in a duplex system (an assay for two analytes), the contribution from each probe can be recovered from a droplet that contains both of the analytes, based on the increased intensity from such a droplet relative to droplets containing only a single analyte. *See id.* And, in any event, the information obtained from the single-analyte droplets is sufficient for Larson's purposes; indeed, this is why Larson expressly teaches to exclude analyzing multi-analyte droplets, as discussed below.

Reply at 15 (citing EX1044 at 159:20-160:14, 160:14-161:14)

EX2018 Struhl Decl. ¶ 61

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Confirms that Larson teaches analysis of droplets with multiple targets

Q. You are discussing the situation where Larson discusses the term of "10 percent occupancy"

A. Right.

Q. And this is the situation where 1 percent of the droplets are two targets?

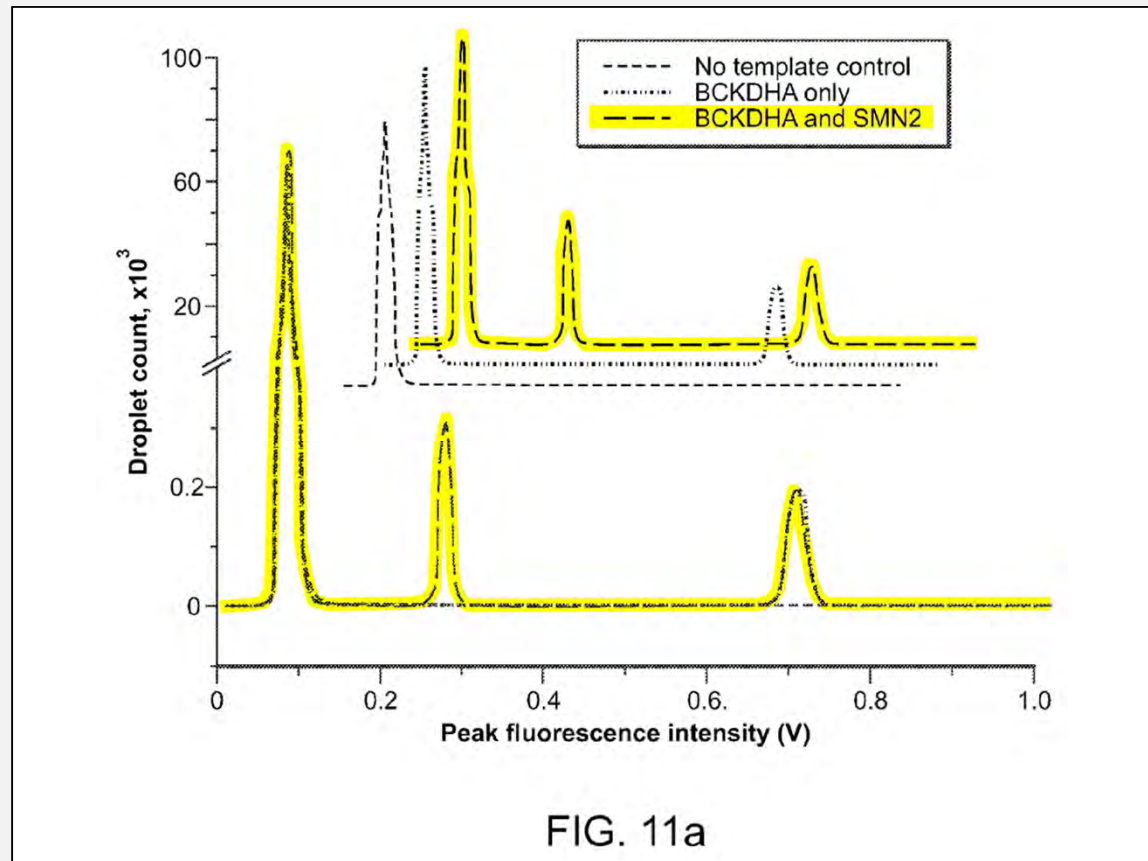
A. That's true.

Reply at 15

EX1044, Struhl Depo. 160:19-24

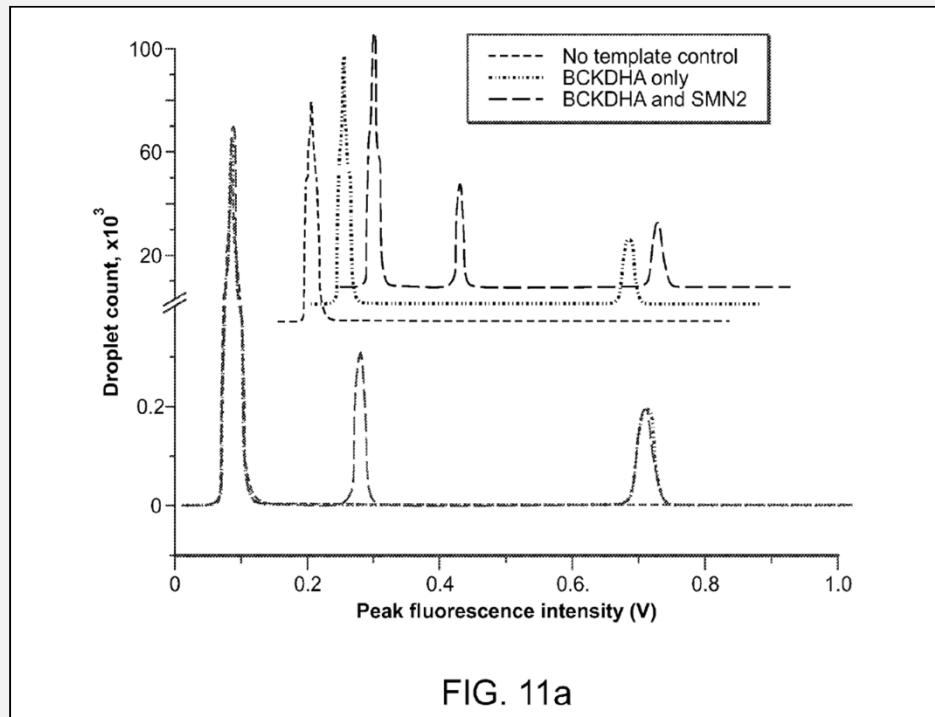
Larson

Teaches the detection of multiple targets within a single droplet



Larson

Teaches the detection of multiple targets within a single droplet



Reply at 15-16

EX1017 Larson, Fig. 11a

[0131] Methods of the invention also include analytical techniques for identification of fluorescence signatures unique to each probe. In this example of the invention, histograms of the droplet fluorescence intensities are shown in FIG. 11a for three different template DNA samples: a no template control (dotted line), BCKDHA only (solid line), and 1:1 BCKDHA to SMN2 (dashed line). For clarity, the histograms are shown both overlapped to highlight the similarity for certain peaks, and offset from each other to reveal all of the features. In the case of 1:1 BCKDHA to SMN2, three populations were readily apparent: a dominant feature appeared at 0.08 V, and two smaller peaks were evident at 0.27 and 0.71 V. The dominant feature at 0.08 V was assigned to PCR(-) droplets since both small peaks disappeared, but the large one remained, in the no template control. The peak at 0.71 V was assigned to BCKDHA since it was the sole feature arising with the addition of just BCKDHA, and the peak at 0.27 V appeared on subsequent addition of SMN2, completing the assignments. A very small peak appeared at ~0.9 V, not visible on the scale of FIG. 11a, that corresponded to droplets occupied by both genes. As another method of the invention, once the different peaks are identified, droplets within each peak were counted corresponding to each possible state (PCR(+)) for either BCKDHA or SMN2, or both, or PCR(-)), and the gene copy number was then determined from the ratio of occupancies. Gene copy numbers for each sample in the serial dilution are plotted in FIG. 11b against expected values (observed ratios of SMN2 to BCKDHA to expected ratios of SMN2 to BSKDHA), with an excellent linear fit ($y=1.01x$) across the full range ($R^2=0.9997$, slope=1.01), demonstrating accurate and precise measurement of the equivalent of 0 to 16 copies of SMN2 per cell.

Reply at 15

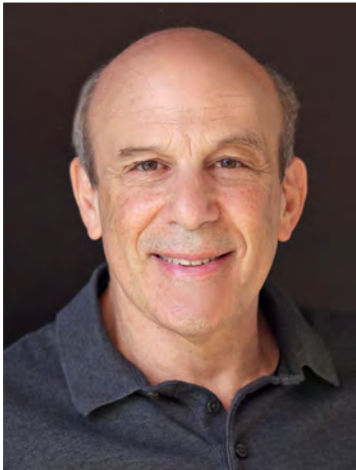
EX1017 Larson ¶ 131

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Struhl

Provides no persuasive response to Figure 11



PO's Expert:
Kevin Struhl PhD

- Q. Okay. Is there any way in which the experiment to Figure 11 doesn't -- is there any element of Claim 1 of the '921 patent that is not part of Figure 11?
- A. The patent, Larson patent, goes out of its way in multiple times to avoid this, and **it does say that you might be able to detect a situation like this**, but it goes specifically out of its way to say that you generally can't do that. And so it's -- it's a totally different thing, and that's on top of the fact that this is a pathetic experiment. I mean, I can't believe they didn't do the control of the M whatever it is by itself and saying that there is a band somewhere or a thing somewhere where you can't see it, what kind of stuff is that? **How do we know they didn't make it up?** I'm not accusing them of being dishonest, but that's not the point. **The point is that you have to provide evidence. There is no evidence here.**

Reply at 16

EX1044, Struhl Depo. 154:6-155:1

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Patent Owner's Response

Contends that Larson excludes multiple occupancy droplets

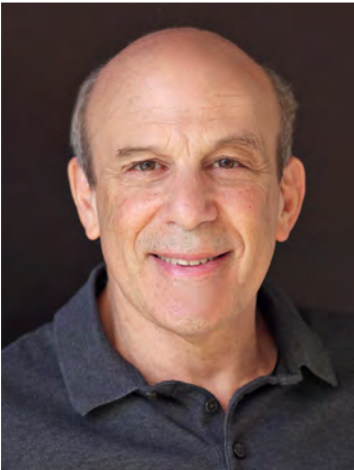
To the extent any droplets nonetheless contain more than one analyte molecule before and during PCR, Larson expressly teaches *excluding* any such droplets from analysis after PCR, but before analyzing the droplets. EX2018, ¶64. More specifically, Larson states “[m]ethods of the invention are able to detect which droplets contain a *heterogeneous population* of molecules and are able to *exclude* those droplets from the analysis.”³ EX1017, ¶172; EX2018, ¶55. Larson

Reply at 17

POR at 19

Dr. Struhl

Relies upon Larson's disclosure regarding polymerase error



PO's Expert:
Kevin Struhl PhD

64. To the extent that any droplets nonetheless contain more than one analyte molecule before and during PCR, Larson expressly teaches excluding any such droplets from analysis after PCR, and before analyzing the droplets. More specifically, Larson states that “[m]ethods of the invention are able to detect which droplets contain a *heterogeneous population of molecules* and are able to *exclude* those droplets from the analysis.” EX1017(Larson), ¶172. Below, I will address each of the emphasized terms in turn.

EX2018 ¶ 64

Larson

PO relies upon ¶ 172 of Larson

[0172] Methods of the invention are able to detect which droplets contain a heterogeneous population of molecules and are able to exclude those droplets from analysis. As droplets containing amplified product flow in a channel through the detector module, the module is able to detect the fluorescent emission in each droplet. Droplets that produce only a single signal are classified as droplets that contain a homogeneous population of target. Since probes that hybridize to the wild-type of the target have a different fluorophore attached than probes that hybridize to a variant of the wild-type of the target, methods of the invention can classify each droplet as containing either a homogeneous population of amplicons of the target or a homogeneous population of amplicons of the variant of the target.

Reply at 17

EX1017 Larson ¶ 172

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Larson

PO omits context: ¶ 172 of Larson is about polymerase error

Polymerase Error Correction

[0171] A polymerase error that converts a wild-type of the target to a variant of the target during an early round of amplification results in a heterogeneous population of target and target variant in a droplet, and may lead to a droplet being incorrectly identified as containing a variant of the target, i.e., a false positive. Such false positives greatly impact the validity and precision of digital PCR results.

[0172] Methods of the invention are able to detect which droplets contain a heterogeneous population of molecules and are able to exclude those droplets from analysis. As droplets containing amplified product flow in a channel through the detector module, the module is able to detect the fluorescent emission in each droplet. Droplets that produce only a single signal are classified as droplets that contain a homogeneous population of target. Since probes that hybridize to the wild-type of the target have a different fluorophore attached than probes that hybridize to a variant of the wild-type of the target, methods of the invention can classify each droplet as containing either a homogeneous population of amplicons of the target or a homogeneous population of amplicons of the variant of the target.

Larson

Discloses that droplets with heterogeneous mixture of targets result from polymerase error

Polymerase Error Correction

[0168] For applications requiring very high sensitivity, such as searching for rare mutations amidst an abundance of wild-type DNA, false positive results can arise from errors from the DNA polymerase itself. For example, during one of the early thermal cycles the polymerase might synthesize the mutant strand of DNA from a wild-type template. This type of error is most likely to occur when the difference between the mutant and the wild-type is very small, such as single nucleotide polymorphism (SNP). In this method of the invention, each droplet contains only a single target nucleic acid, if any at all. In the preferred embodiment, this is accomplished under the conditions of terminal dilution. Droplets that contain amplification products that are a wild-type of the target are detected based on emission from the fluorophore that is released from the probe that hybridizes to the wild-type of the target. Droplets that contain the variant of the target are detected based on emission from the fluorophore that is released from the probe that hybridizes to the variant of the target. Since each droplet starts with only a single nucleic acid molecule, the resultant amplification products in each droplet are either homogeneous for the target or homogenous for the variant of the target.

[0169] However, certain droplets will contain a heterogeneous mixture of both target and target variant due to polymerase errors during the PCR reaction. Error rates in PCR

Larson

Droplets excluded where polymerase error is an issue

[0015] The invention further relates to methods for analyzing a target nucleic acid. More particularly, methods of the invention are able to detect polymerase errors that occur during a PCR reaction and are able to exclude from analysis amplification products that are a result of a polymerase error. Methods of the invention are particularly useful in digital PCR where a polymerase error may result in a partitioned section of sample being incorrectly identified as containing a mutant allele, i.e., a false positive. Such false positives greatly impact the validity and precision of digital PCR results. Methods of the invention are able to uniquely detect multiple targets with the same optical color. Methods of the invention are particularly useful in digital PCR where it is desirable to identify multiple different target molecules that may be present in the starting test fluid.

Petition at 23; 29; Reply at 17

EX1017 Larson ¶ 15

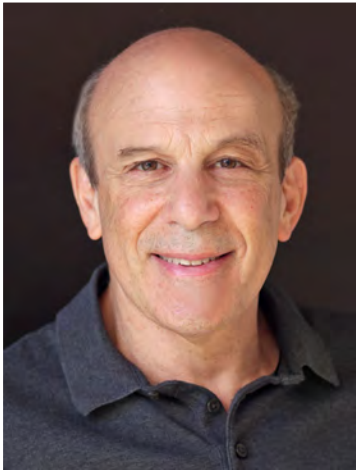
Larson

Droplets with “only the variant” are indicative of disease

[0018] Once droplets containing a heterogeneous population of amplicons are excluded, droplets that contain a homogeneous population of amplicons are analyzed. Any analytical technique known in the art may be used. In certain embodiments, analyzing the droplets involves determining a number of droplets that contain only wild-type target, and determining a number of droplets that contain only a variant of the target. Generally, the presence of droplets containing only the variant is indicative of a disease, such as cancer. The variant may be an allelic variant. An exemplary allelic variant is a single nucleotide polymorphism. The variant may also be a specific haplotype. Haplotypes refer to the presence of two or more variants on the same nucleic acid strand. Haplotypes can be more informative or predictive than genotypes when used to determine such things as the presence or severity of disease, response to drug therapy or drug resistance of bacterial or viral infections. Because each droplet contains only one template strand it is an ideal vessel for the determination of haplotypes. The detection of two or more variants in a single droplet that contains a single intact nucleic acid strand identifies the haplotype of the variants on that strand. The presence of two or more markers in the same droplet can be identified by such methods as the presence of dyes of multiple colors or the increase in the intensity of a single dye or a combination of both. Any method that allows the identification of multiple variants in a single droplet enables the determination of a sample's haplotype.

Dr. Struhl

Has no understanding of why polymerase error is relevant



PO's Expert:
Kevin Struhl PhD

Q. Okay. And do you have any idea where the variant is going to come from in those droplets that have both the wild type and the variant?

A. I'm not sure what they are getting at, but they -- I think they are getting at an error that happens during PCR. I'm not sure why that's relevant to anything.

Reply at 18

EX1044, Struhl Depo. 120:17-20 (objections omitted)

A. I want to just emphasize that last point. This is outside the scope of my testimony, and my declaration, and outside for a reason. I didn't think this was worth talking about, that polymerase error rate is really low and even says it what it is here. It's incredibly low. And so even if you had an occasional bad droplet somewhere, it wouldn't matter. And never even dawned on me why this is relevant to anything, and why I didn't opine on it, and I'm not going to sit here and opine on it now.

Reply at 18

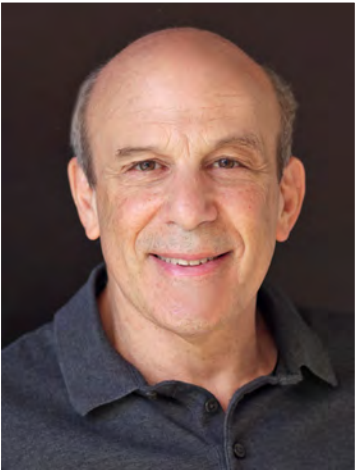
EX1044, Struhl Depo. 127:3-12

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Struhl

Has no understanding of why polymerase error is relevant



PO's Expert:
Kevin Struhl PhD

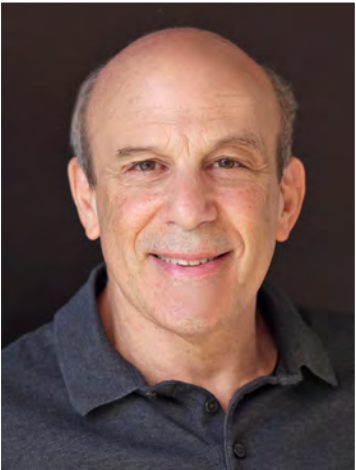
- Q. I was asking something different. The section in Paragraphs 168 to 173, you don't recall opining on any of these paragraphs?
- A. Off the top of my head, I don't remember opining on it. I mean, anything relating to polymerase error, I don't remember, recall opining upon. And as I said, I don't think it's very important, which is why I didn't opine on it.

Reply at 18

EX1044, Struhl Depo. 127:20-128:3 (objections omitted)

Dr. Struhl

Has no understanding of why polymerase error is relevant

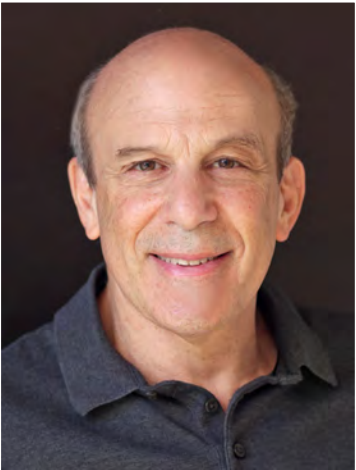


PO's Expert:
Kevin Struhl PhD

- Q. And that's what Paragraph 15 is talking about, a situation where the polymerase makes an error and then introduces an error, correct?
- A. It's about -- the paragraph, it's about polymerase errors, yes. I can read it, yes.
- Q. And what happens, you get the error, and the error looks like it's a mutant allele when it's really just an error that was introduced from a normal wild type; is that fair?
- A. An error just means when you copy something that you didn't copy it faithfully. It's actually quite uncommon.
- Q. Okay. But this is talking about -- it's talking about false positives, right?
- A. I must say, I don't really understand why this paragraph is here. I mean, I don't know why there is such a big emphasis on this.
- Q. You don't understand why it's there?
- A. I don't understand why they are making a big deal out of something that is not a big deal.

Dr. Struhl

Has no understanding of why polymerase error is relevant



PO's Expert:
Kevin Struhl PhD

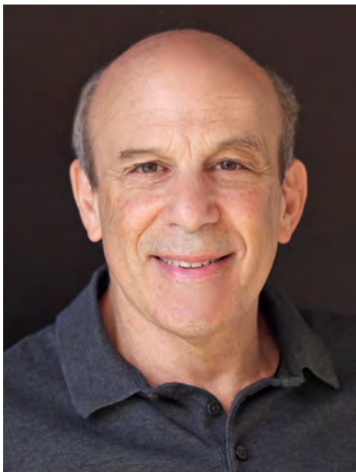
- Q.** Okay. And then the patent goes on and talks about how you can exclude those droplets that have the normal and then the PCR error together; is that fair?
- A.** It says it, but I didn't really pay attention; that's why it's not on my -- it's not in my declaration because I don't -- I didn't pay attention to that because I don't really think it's much of a problem.

Reply at 18

EX1044, Struhl Depo. 121:1-9

Dr. Struhl

Has no understanding of why polymerase error is relevant



PO's Expert:
Kevin Struhl PhD

- Q. All right. Now, Paragraph 18, it states -- I'm sorry Paragraph 18. It states, "Generally, the presence of droplets containing only the variant is indicative of a disease, such as cancer." Do you see that?
- A. Yes. That's sloppy. That's sloppy English.
- Q. Okay. Now, why wouldn't the droplets that have both the wild type, and the variant be indicative of cancer?
- A. I don't get what you are saying. They're a variant if it's a real allele. That's a real allele, okay. That could be predictive of cancer. That has nothing to do with this.

Reply at 18

EX1044, Struhl Depo. 122:25-123:16 (objections omitted)

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Petitioner's Expert

PO mischaracterizes Dr. Batt's testimony



Petitioner's Expert:
Carl Batt PhD

2. **Larson Does Not Teach Discarding All Droplets with Multiple Occupancy**

24. Dr. Struhl mischaracterizes Larson by opining that “[t]o the extent that any droplets nonetheless contain more than one analyte molecule before and during PCR, Larson expressly teaches excluding any such droplets from analysis after PCR, and before analyzing the droplets.” EX2018 at ¶ 64.

25. **Larson teaches exclusion of *amplification products resulting from polymerase errors during PCR*, not droplets containing multiple targets resulting from the Poisson distribution under limiting dilution.** Larson teaches that “[f]or applications requiring very high sensitivity” (EX1017 at ¶ 168)—such as the

PO Sur-reply at 5-6

EX1043 Batt Decl. ¶¶ 24-25

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Petitioner's Expert

Figure 9 of Larson teaches unambiguous detection of all combinations of three targets



Petitioner's Expert:
Carl Batt PhD

336. A POSA would have readily understood, detected, and distinguished these signals from one another. Specifically, because the slightly less than 0.2-intensity signal is not multipliable into the slightly more than 0.4-intensity signal, even a multiple of intensity 3 could be distinguished from intensity 1 because the various combinations of analytes would result in distinguishable signals for the three analytes. The signals from P1, P2, P3, P1+P2, P1+P3, P2+P3, and P1+P2+P3 have different intensities as the combinations are additive and would each sum to unique values in each of the two colors used. EX1017 ¶ 121, Fig. 9C. As discussed, this unique sum of values for the different analytes provides a method for a POSA to use the assays in Larson to detect multiple analytes in one droplet.

Petition at 56-57

EX1002 Batt Decl. ¶ 336

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Larson

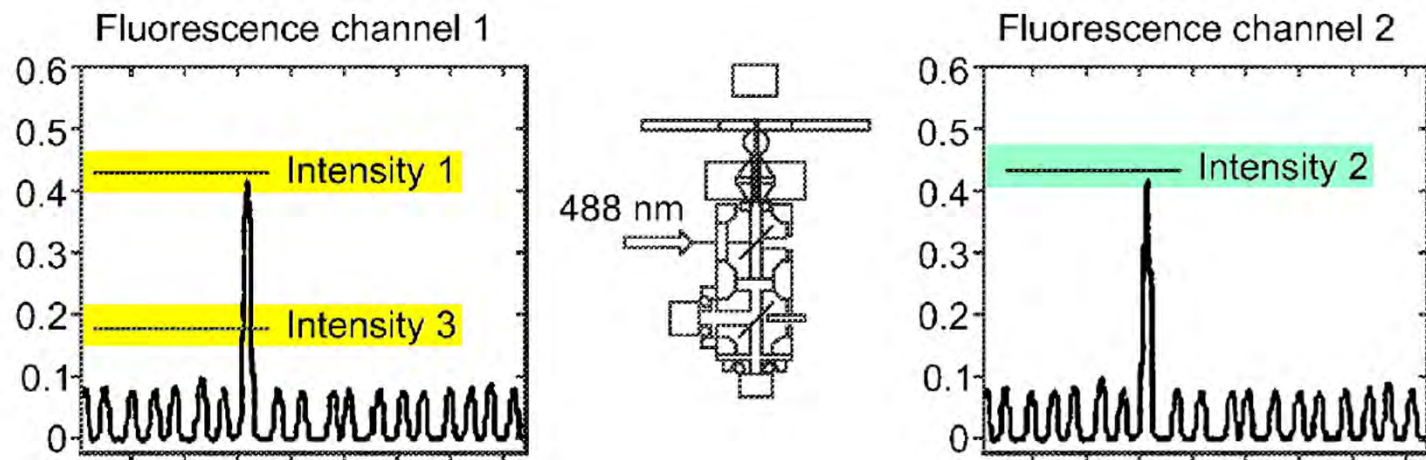
Figure 9 permits unambiguous detection of all combinations of targets

[0121] Methods of the invention involve performing accurate quantitation of multiple different DNA targets by dPCR using probes with the same fluorophore. FIG. 9 is a schematic depicting two-color detection of three genetic sequences with a microfluidic device. As shown in Panel A of FIG. 9, a template DNA is amplified with three sets of primers: forward primers (F1, F2 and F3) and reverse primers (R1, R2 and R3). Probes (P1, P2 and P3) are labeled with fluorophores (color 1, color 2 and color 1) and bind to the target genetic sequences (target 1, target 2 and target 3) (Panels B and C). Microdroplets are made of diluted solution of template DNA under conditions of limiting or terminal dilution. Microdroplets containing target sequence 1 or 3 emit fluorescence of color 1 at two different intensities; and microdroplets containing target sequence 2 emit fluorescence of color 2. The number of microdroplets containing target 1, 2 or 3 is shown by histogram in Panel D.

Larson

Figure 9 permits unambiguous detection of all combinations of targets

FIG. 9C



Petition at 55

EX1017, Fig. 9C

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Batt

The testimony cited by PO does not support its position



Petitioner's Expert:
Carl Batt PhD

- Q. You didn't account for any of the reasons why it might not work, correct?
- A. Right, **because the overwhelming thing is that it would work.** That's a reasonable expectation. With somebody reading this of ordinary skill in the art, they may say it might not work, it might work, but they are going to come away from this with a reasonable expectation of success.

PO Sur Reply at 1

EX2017, Batt Depo. 185:6-15

Dr. Batt

The testimony cited by PO does not support its position



Petitioner's Expert:
Carl Batt PhD

- Q.** And those are issues that Larson avoids by having only one template molecule per droplet, that's what he says in paragraph 100, correct?
- A.** So if you are looking at that phrase that says these issues are avoided in droplet PCR, that is correct. And it says there is only one. In other words, you can't exclude only one a priori, but that would be your aspiration.

PO Sur Reply at 3

EX2017, Batt Depo. 129:13-22

Dr. Batt

The testimony cited by PO does not support its position



Petitioner's Expert:
Carl Batt PhD

- Q.** And he didn't describe any of those droplets in the figures, did he?
- A.** No, but he talks about – you know, he does talk about multiple occupancy in various points of the patent and does discuss them. And again, I will say on a statistical basis those droplets exist.

PO Sur Reply at 3

EX2017, Batt Depo. 145:14-20

Dr. Batt

PO takes Dr. Batt's testimony out of context



Petitioner's Expert:
Carl Batt PhD

- Q. when you have higher degrees of multiplexing, and one reason for that is you want to reduce cross reactivity that is unintended; is that correct?
- A. That is correct, yes. But it does gone on to say droplets would be double resulting in at least as high or higher than the -- so it does account for multiple occupancy. It does talk about the outcomes. It does say for a simple duplex system the contribution for each probe could be recovered.

PO Sur Reply at 5 (context for citation to 168:21-169:23)

EX2017, Batt Depo. 167:25-168:13

Dr. Batt

PO takes Dr. Batt's testimony out of context



Petitioner's Expert:
Carl Batt PhD

- Q.** And he doesn't say anything about microdroplets containing targets 1 and 3, correct?
- A.** Again, I will come back and repeat what I just said, it doesn't mean that they don't exist. He is simply focusing, as you do in these analyses, focusing in on the populations of those particular singular events. But it doesn't mean that there isn't droplets that contain 1, 2 and 3, 1 and 2, 1 and 3, 2 and 3.
- Q.** But you can't tell from figure 9D 20 whether those droplets exist?
- A.** So I do not have powerful enough eyes to look at this histogram and find individual droplets.

PO Sur Reply at 14

EX2017, Batt Depo. 115:8-18

Ground 3: Obviousness In View Of Larson & Saxonov

Patent Owner's Response

Expressly declined to respond to grounds based on Saxonov

The '921 Patent claims are novel and patentable over Larson, whether alone or combined with Silverbrook. While Petitioner relies on Saxonov for Grounds 3 and 4, the Board found that the Petition fails to establish Saxonov qualifies as prior art. Paper 11, 28-29. Accordingly, and for the reasons PO discussed in its POPR (Paper 8, 29-42) the portions of Grounds 3 and 4 that rely upon Saxonov necessarily fail and PO does not further address them herein. *See also supra*, §II.D.

Reply at 13

POR at 27

The Board's Institution Decision

Made only a “preliminary” determination on whether Saxonov is prior art

3. *Prior Art Status of Saxonov*

The parties disagree as to whether Saxonov is available as prior art under 35 U.S.C. 102(e). We address this contention here to provide the parties with our preliminary determination regarding this disputed issue. However, as noted below in the discussion of this ground, at this preliminary stage, we focus on Petitioner's contention that the challenged claims are rendered obvious by Larson alone.

Thus, because Petitioner has not shown sufficiently that the Petition explains how each of the relied-upon disclosures in Saxonov Figure 10, and Saxonov paragraphs 18 and 78–79, which are not present in the '013 provisional, are supported by the '013 provisional, we determine that Petitioner has not shown that those cited portions of Saxonov are entitled to the priority date of the '013 provisional. Accordingly, on this record, we preliminarily determine that those relied-upon portions of Saxonov do not qualify as prior art to the '921 patent.

Scheduling Order

Arguments omitted from POR may be deemed waived

a. A response to the petition (37 C.F.R. § 42.120). If Patent Owner elects not to file a response, Patent Owner must arrange a conference call with the parties and the Board. Patent Owner is cautioned that any arguments not raised in the response may be deemed waived.

Scheduling Order at 10

Law

Failure to respond can lead to waiver

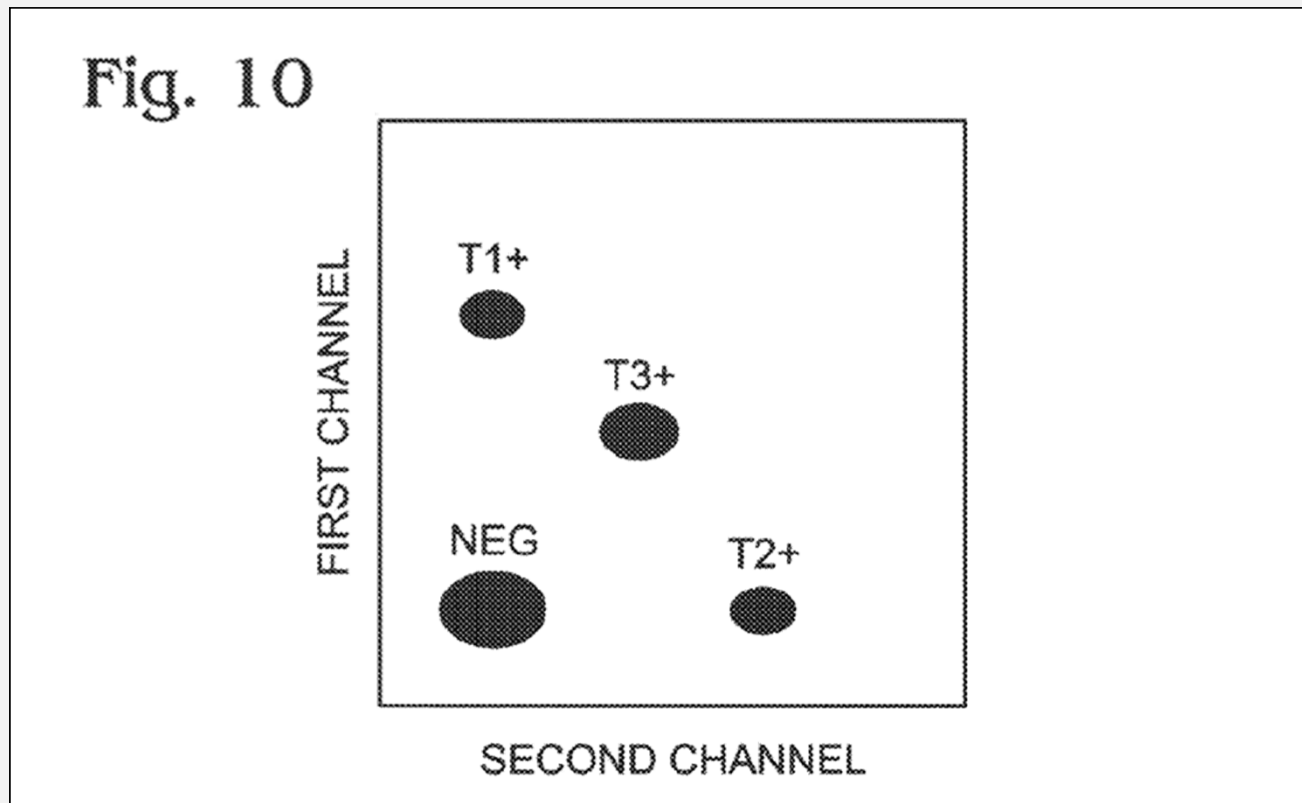


“To the extent PO has not responded to Petitioner’s arguments and evidence with respect to this claim limitation, we find that PO has waived any such argument.”

Microsoft Corp. v. IPA Techs., Inc., No. IPR2019-00813, 2020 WL 6084149, at *67 (P.T.A.B. Oct. 15, 2020)

Saxonov

Discloses unambiguous detection of different combinations of targets in droplets



Petition at 59

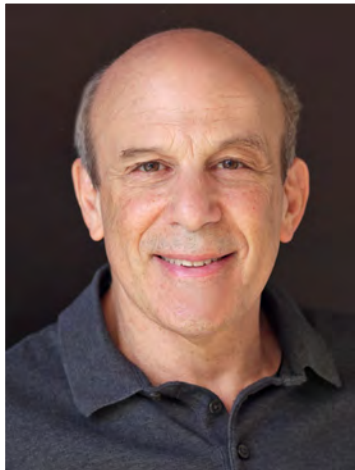
EX1004 Saxonov, Fig. 10

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Struhl

Agrees that vast majority of Saxonov is carried over from provisional



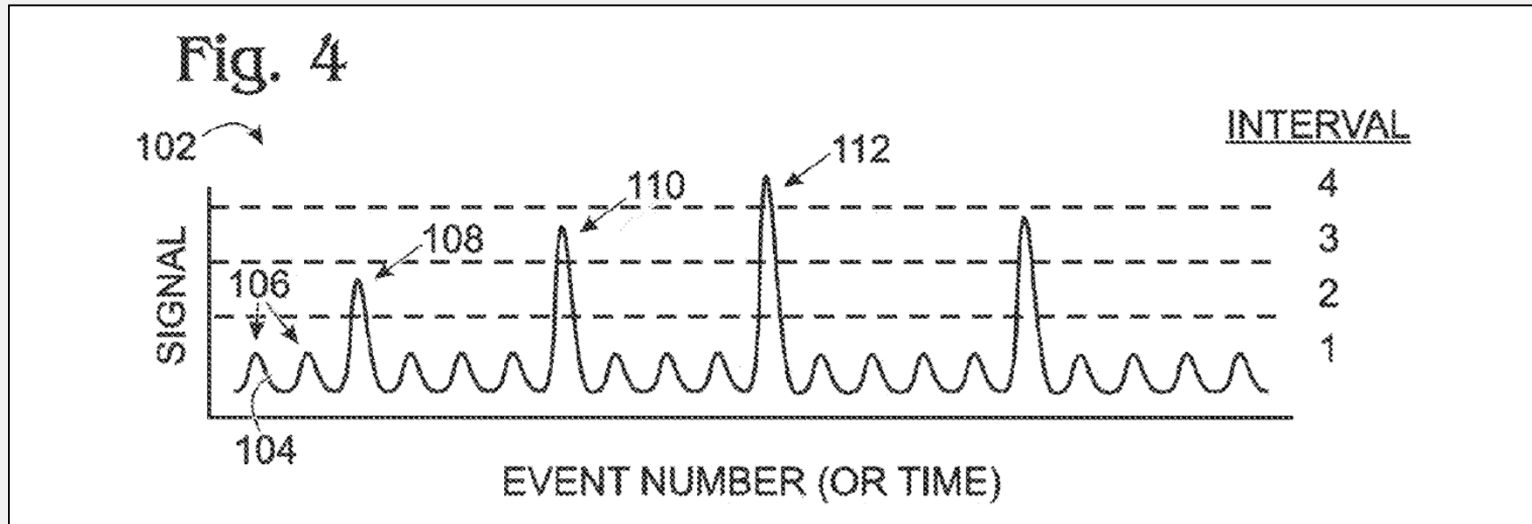
PO's Expert:
Kevin Struhl PhD

88. For example, FIGS. 1-8A and 8B of Saxonov generally correspond to, and are carried over from, FIGS. 1-8 of the Saxonov Provisional, with certain differences. However, FIGS. 9 and 10 of Saxonov are *absent* in the Saxonov Provisional. Nor does any disclosure in the Saxonov Provisional, as it would have been understood by a POSA, support or disclose the subject matter relied on by Petitioner from these figures. *See* EX2007, 3, 16-17.

89. As another example, ¶¶2-16, 19-36, 38-53, 45-68, and 72-75 of Saxonov generally correspond to page 1, line 10 through page 20, line 22 of the Saxonov Provisional. *Id.*; *see also* EX2007. However, ¶¶17, 18, 37, 44, 69-71, and 76-80 of Saxonov contain significant new subject matter which is *not found in, or supported by*, the Saxonov Provisional. *Id.*; *see also* EX2007.

Saxonov Provisional

Teaches detecting multiple targets in a single droplet



Reply at 21

EX1004 Saxonov, Fig. 4

and flowing through an examination region of the channel. The signal may be analyzed to determine whether neither Target, Target 1 alone, Target 2 alone, or both Targets 1 and 2 are present in each droplet. In particular, the strength or intensity of the signal in a

Reply at 21

EX1007.021 Saxonov Provl 15:17-19

Saxonov Provisional

Discloses eight clusters of data corresponding to droplets with multiple targets

The assays may be extended in various ways. In some embodiments, the assays may involve analysis of more than two targets in the same channel. For example, an assay for three targets would generate eight clusters or populations of data, separated by intensity. In the same or other embodiments, some targets may be analyzed in one channel (e.g., a FAM channel), and other targets may be analyzed in other channels (e.g., a VIC channel). Three targets, two in a first channel and one in a second channel, would again generate eight clusters or populations of data, but they would be separated in a two-dimensional intensity space and so in principle more easily resolvable.

Reply at 20

EX1007.011-012 Saxonov Provl 5:19-6:3

Saxonov Provisional

Discloses eight clusters of data corresponding to droplets with multiple targets

	Target A	Target B	Target C	Intensity
Population 8	+	+	+	Highest
Population 7	+	+	-	Intermediate
Population 6	+	-	+	Intermediate
Population 5	-	+	+	Intermediate
Population 4	+	-	-	Intermediate
Population 3	-	+	-	Intermediate
Population 2	-	-	+	Intermediate
Population 1	-	-	-	Lowest

Reply at 20

EX1007.017 Saxonov Provl 11:3

Saxonov

Saxonov ¶ 20 teaches eight populations of droplets, just like the provisional

[0020] The assays may be extended in various ways. In some embodiments, the assays may involve analysis of more than two targets in the same channel. For example, an assay for three targets may generate eight clusters or populations of data, separated by intensity. In the same or other embodiments, some targets may be analyzed in one channel (e.g., a FAM channel), and one or more other targets may be analyzed in one or more other channels (e.g., a VIC channel). Three targets, two in a first channel and one in a second channel, would again generate eight clusters or populations of data, but they would be separated in a two-dimensional intensity space and so in principle more easily resolvable.

Petition at 52

EX1004 Saxonov ¶ 20

Saxonov

Saxonov ¶ 78 teaches eight populations of droplets, just like the provisional

[0078] FIG. 10 schematically represents an exemplary graph showing droplet (or peak) intensities for a T1/T2/T3 droplet assay that may be performed with the droplets of FIG. 9. Data from the T1, T2, and T3 probes are collected in a first optical channel and a second optical channel and plotted as a function of intensity in the first channel (vertical axis) versus intensity in second channel (horizontal axis). Intensities produced by exemplary populations or clusters of negative, T1-positive, T2-positive, and T3-positive droplets are identified by filled ellipses, with the size of each ellipse corresponding to the number of droplets in each population. To simplify the presentation, double-positive populations (T1+/T2+, T1+/T3+, and T2+/T3+) and the triple-positive population (T1+/T2+/T3+) are not shown.

Petition at 59-60

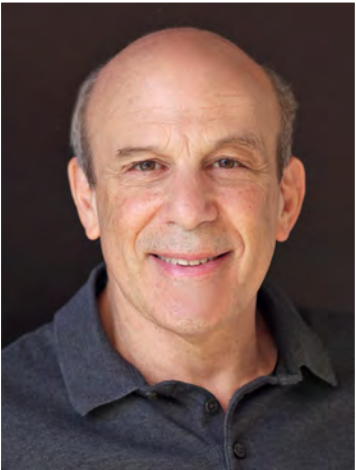
EX1004 Saxonov ¶ 78

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Struhl

Saxonov “essence” is one target per droplet

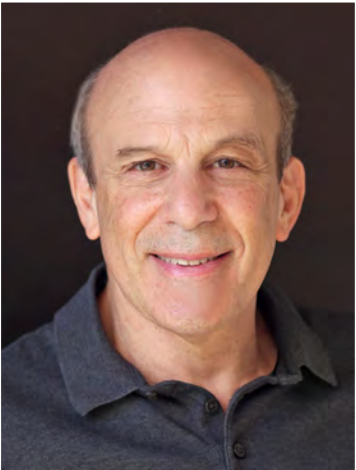


PO's Expert:
Kevin Struhl PhD

- Q. The essence of the Saxonov reference, is it at 1004? Is it one target per droplet?
- A. That's the ideal setup.
- Q. Okay. You would -- you would feel comfortable calling that the essence of Saxonov?
- A. A significant part of it.

Dr. Struhl

Saxonov teaches detecting multiple targets in a single droplet



PO's Expert:
Kevin Struhl PhD

- Q. Okay. All right. Let's go to your original declaration in this case, Exhibit 2001. Let's go to Paragraph 80.
- A. Okay.
- Q. What is being shown in Paragraph – go ahead and review Paragraph 80.
- A. And Figure 4. Figure 4, yes. Yes, okay.
- Q. Okay. What is being shown in Figure 4? Can you describe it for me?
- A. First of all, **Figure 4 is not real data**. So what it actually would look like is anybody's guess, but the way it's portrayed is that you see, you know, **108 is target 1. Let's see. Sorry. 106 is no target. 108 is target 1 but not target 2. 110 is target 2 but not target 1. And 112 is both.**
- Q. **Okay. 112 are droplets that have both targets?**
- A. **Yes.**

Saxonov

Teaches using a Poisson distribution where individual partitions can have more than one target

[0043] A level of each target may be determined, indicated at 52. The level of each target may be determined based on the respective numbers of partitions positive for the target alone and for the target in combination with any other target(s). The calculation may be based on each target having a Poisson distribution among the droplets. The level may be a concentration. The levels may, for example, be estimated based on finding solutions to a series of linear equations. The total number of partitions may be counted or, in some cases, estimated. The partition data further may be used (e.g., directly and/or as concentration data) to estimate copy number (CN) and copy number variation (CNV), using any suitable algorithms such as those described elsewhere in the present disclosure.

Petition at 58

EX1004 Saxonov ¶ 43

Saxonov

Teaches using a Poisson distribution where individual partitions can have more than one target

[0003] Digital assays generally rely on the ability to detect the presence or activity of individual copies of an analyte in a sample. In an exemplary digital assay, a sample is separated into a set of partitions, generally of equal volume, with each containing, on average, less than about one copy of the analyte. If the copies of the analyte are distributed randomly among the partitions, some partitions should contain no copies, others only one copy, and, if the number of partitions is large enough, still others should contain two copies, three copies, and even higher numbers of copies. The probability of finding exactly 0, 1, 2, 3, or more copies in a partition, based on a given average concentration of analyte in the partitions, is described by a Poisson distribution. Conversely, the concentration of analyte in the partitions (and thus in the sample) may be estimated from the probability of finding a given number of copies in a partition.

Dr. Batt

Confirms that PO would be motivated to apply Saxonov to Larson



Petitioner's Expert:
Carl Batt PhD

disclosed through the combination of Larson and Saxonov. Specifically, Larson teaches how to identify three different targets with two colors, based on their difference in intensity, and teaches that multiple targets can be in a single droplet, and which "multiple occupancy" its methods could handle. See *supra* Section XI.A; EX1017 ¶¶ 129, 140-141.

Petition at 58

EX1001 Batt Decl. ¶ 345

Analyte	FAM	HEX
N2	1	0
E	1	1
MHV	0	1

EX1012 at 11

[0140] Another method of the invention involves performing higher-plex assays using multiple different probe colors (i.e. fluorophores), however unlike the strategy above where each target is identified by single type of probe with a unique color and intensity, instead in this method a single target may be identified by multiple probes that constitute a unique signature of both colors and intensities. For example, a single droplet might contain four different probes for measuring three different targets (say, T1, T2, and T3). Two probes might be of color A (say, A1 and A2), and two probes might be of color B (say, B1 and B2). T1 is measured by probe A1, T2 is measured by probe B1, but T3 is measured by both probes A2 and B2. Thus, when a droplet contains T1 only increased fluorescence appears in color A. When a droplet contains T2 only increased fluorescence appears in color B. However when a droplet contains T3, increased fluorescence appears in both colors A and B.

Petition at 25; Reply at 14

EX1017 Larson ¶ 140

Dr. Batt

PO would be motivated to apply Saxonov to Larson



Petitioner's Expert:
Carl Batt PhD

disclosed through the combination of Larson and Saxonov. Specifically, Larson teaches how to identify three different targets with two colors, based on their difference in intensity, and teaches that multiple targets can be in a single droplet, and which "multiple occupancy" its methods could handle. See *supra* Section XI.A; EX1017 ¶¶ 129, 140-141.

Petition at 58

EX1001 Batt Decl. ¶ 345

[0129] Another advantage of the dPCR approach for multiplexed reactions is that the different reactions do not compete with each other for reagents as they would in a bulk qPCR assay. However, the possibility for unintended cross-reactivity remains. A multiplexed assay can require a more dilute sample. For instance, at 10% occupancy a duplex reaction would have double occupancy 1% of the time. Hence 1 in 10 PCR+ droplets would be doubles, resulting in a final intensity at least as high and possibly higher than the brighter of the two probes. For a simple duplex system the contribution from each probe could be recovered. In this example the total number of PCR+ droplets for probe 1 would be (Probe 1)+(Probe1+Probe2). Higher degrees of multiplexing would require greater dilution. For example, for a 4-plex at 1% occupancy the probability of one probe overlapping any of the other 3 is ~3%, and that error may be too high for some applications. The need for large dilutions strongly favors the large number of dPCR reactions.

Petition at 52

EX1017 Larson ¶ 129

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Dr. Batt

PO would have had a reasonable expectation of success

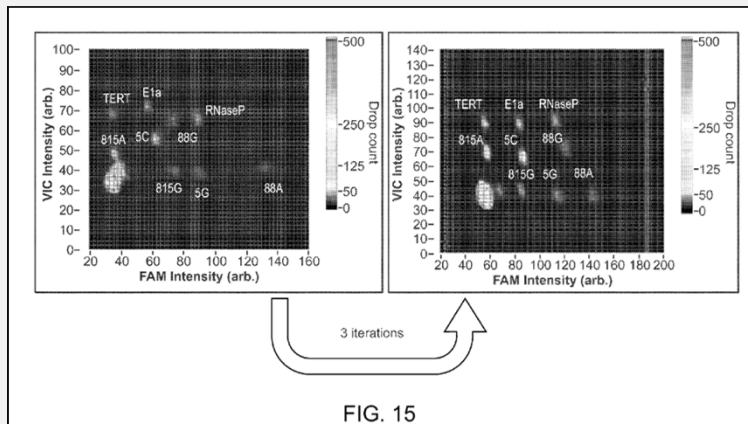


Petitioner's Expert:
Carl Batt PhD

347. Perhaps this goes without saying, but a POSA would have had a reasonable expectation of success in achieving these results, since Larson and Saxonov teach the same assay. But, more specifically, Larson teaches, as discussed, tuning the intensity of the probes that correspond to a target in order to optimize the probe's detection. See *supra* Sections IX.A.2, XI.A, K; EX1017 ¶¶ 134-137, 146.

Petition at 58

EX1043 Batt Decl. ¶ 347



Petition at 45-46

FIG. 15

EX1017 Larson Fig. 15

BIO-RAD

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

Secondary Considerations

Law

Weak secondary considerations do not overcome strong obviousness case

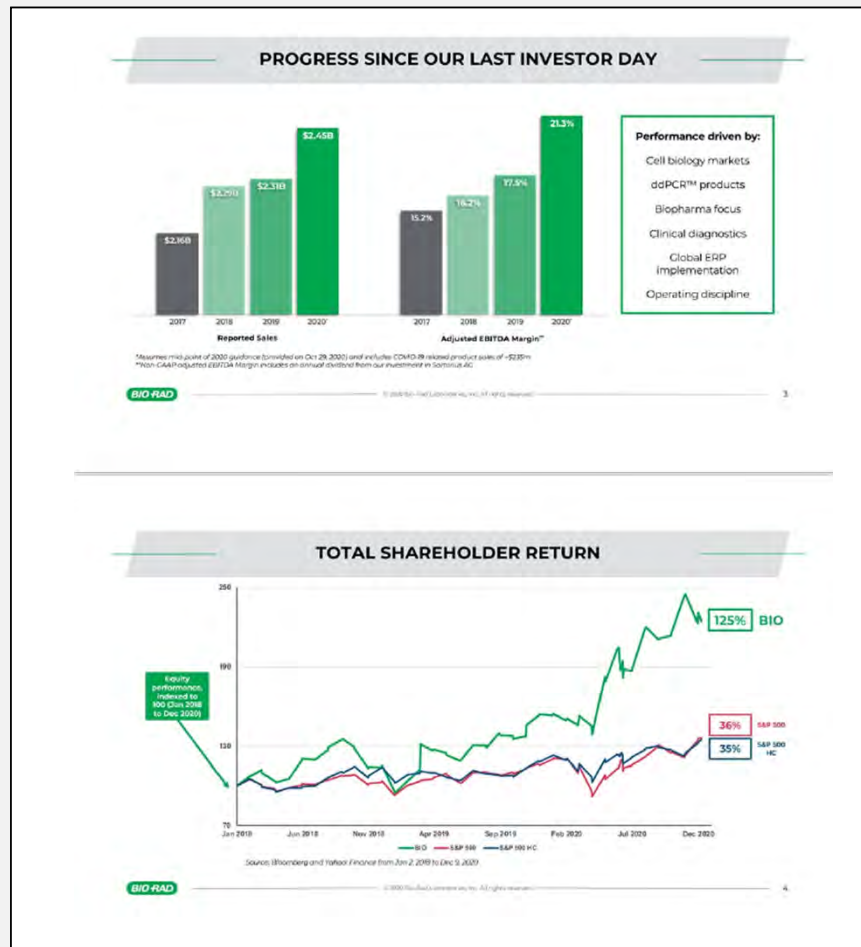


“Moreover, weak secondary considerations generally do not overcome a strong prima facie case of obviousness.”

W. Union Co. v. MoneyGram Payment Sys., Inc., 626 F.3d 1361, 1373 (Fed. Cir. 2010)

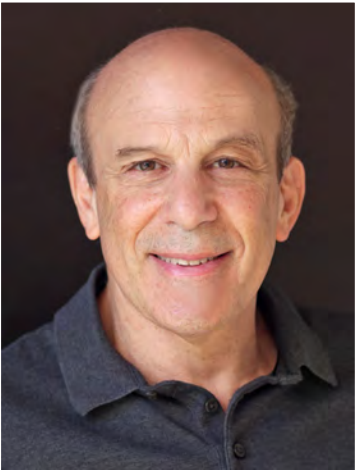
Secondary Considerations

PO's evidence of alleged commercial success



EX2021 at 3

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Cannot credibly opine on long felt need

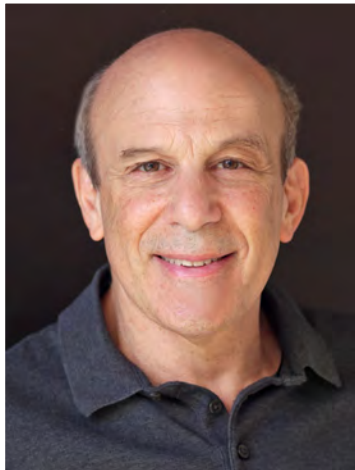
"I did not do any historical analysis for anything."

Reply at 4

EX1044, Struhl Depo. 181:6-9

Dr. Struhl's Testimony Should Be Accorded No Weight

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

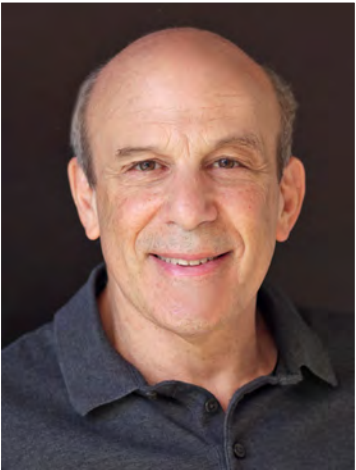
Frequently cites the "Petition"

17. Petitioner asserts that a POSA relevant to the '921 Patent as of February 3, 2012 would have a Ph.D. in molecular biology, genetics, biochemistry, or a related discipline, with at least a year of experience in quantitative and/or digital PCR and fluorescence-based analyte detection. **Petition, 9-10.** Alternatively, the Petitioner asserts that a POSA would have been someone with greater relevant experience sufficient to compensate for having less relevant formal education. *Id.* The Petitioner states that a POSA would have "understood" several technologies and techniques which are enumerated in the Petition. *Id.*

EX2018 Struhl Decl. ¶ 17

Dr. Struhl

Did not understand his citations to the "Petition"



PO's Expert:
Kevin Struhl PhD

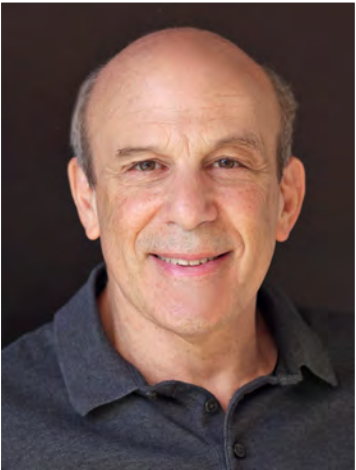
- Q. In reviewing your declarations to prepare for your deposition testimony, did you ever come across your citations to petition and ask yourself: Hey, I wonder what that is maybe I should find out what it is I'm citing?
- A. I didn't. There is so many places of citing. I -- when I was writing it, I -- when we were writing it, you know, I would look at the citation, but that was ages ago.
- Q. Okay. And you don't recall what it was when you were helping to write this, that you were citing to when you cited the petition in Paragraph 17?
- A. I said at the time when I'm citing something, I looked; but now I don't remember specifically what it is. That's why I can say, "This looks familiar in a general sense," but I can't swear that's exactly the same thing.

Reply at 6

EX1044, Struhl Depo. 22:20-23:12 (objections omitted)

Dr. Struhl

Did not recognize the "Petition"



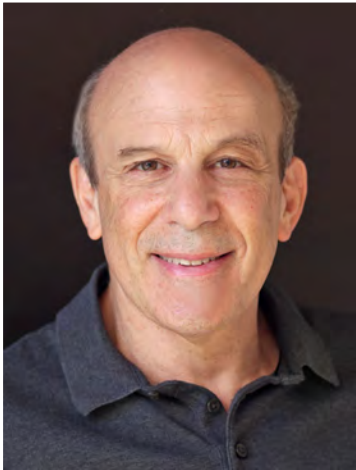
PO's Expert:
Kevin Struhl PhD

- Q. Just go ahead, and let me know if this document that is entitled, "Petition for Inter Partes Review of U.S. Patent Number 11,829,921," is something you think you've looked at before. And if you can't answer that question, let me know. If you don't know, let me know that as well.
- A. Well, it looks familiar, but I don't see it on my list, so I don't really know how to answer the question.

Reply at 6

EX1044, Struhl Depo. 19:24-20:7

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Did not recognize the "Petition"

Q. Okay. And just to be clear, sitting here today, you don't recall what it is you are referring to when you cited petition in your declaration Exhibit 2018?

A. Right now, I don't remember. I don't remember every citation as to what exactly it was being cited to.

Reply at 6

EX1044, Struhl Depo. 24:17-23

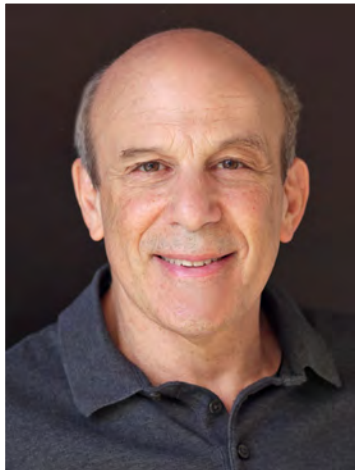
Q. ... Is the definition for the level of ordinary skill in the art that appears on pages 9 and 10 of Exhibit 1043, the Petitioner's definition of the level of ordinary skill in the art that you reviewed and adopted for the purposes of your opinions in this case?

A. I think I stated where I got my thing from, the Petitioner, and so it could have been this. I am a little bit confused. I don't understand what you are asking.

Reply at 6

EX1044, Struhl Depo. 30:6-14

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Frequently invokes the concept of “priority date”

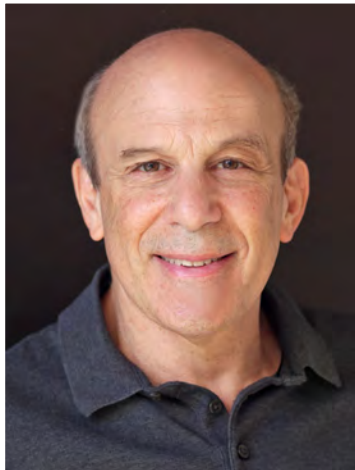
E. Priority Date

26. I understand that to be entitled a priority date of an earlier-filed application, the earlier-filed application must provide support for the invention claimed in the later-filed application as required by the “written description” requirements of 35 U.S.C. § 112. I understand that an adequate written description requires showing that the inventor was in possession of the invention at the time of the patent application’s filing.

EX2018 Struhl Decl. ¶ 26

Dr. Struhl

Frequently invokes the concept of “priority date”



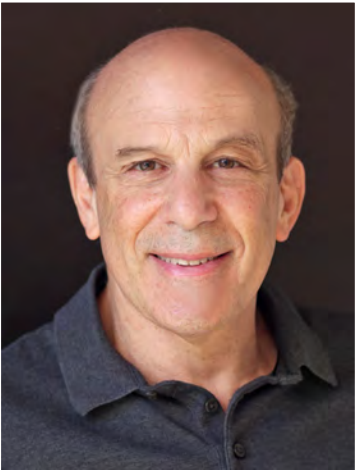
PO's Expert:
Kevin Struhl PhD

D. Obviousness

23. I understand that a patent claim also, or alternatively, is unpatentable if it would have been obvious to a POSA during the relevant timeframe. I understand that the following factors must be evaluated to determine whether the claimed subject matter was obvious: (1) the scope and content of the prior art; (2) the difference or differences, if any, between the subject matter defined by the claim of the patent under consideration and what is disclosed in the prior art; (3) the level of ordinary skill in the relevant field at the time of the patent's earliest priority date or, possibly, at an earlier time; and (4) so-called “objective evidence of non-obviousness.” However, I understand that, for the present, I can ignore both the possibility that obviousness is to be determined at a date earlier than the patent's earliest priority date and the possibility that Patent Owner will submit objective evidence of non-obviousness.

Dr. Struhl

Did not know or understand relevance of priority date



PO's Expert:
Kevin Struhl PhD

Q. What is the priority date for the '921 patent?

A. I told you, I don't know what date it is. I'm not a lawyer. I don't -- that's a legal question.

Q. Did the lawyers tell you what the priority date should be?

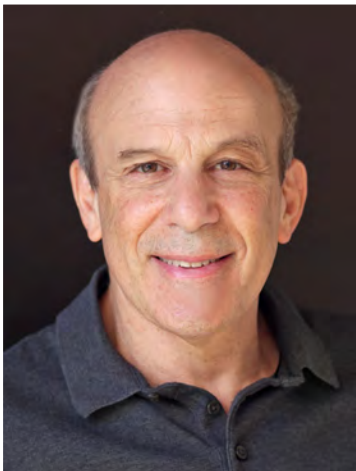
A. I don't remember. Maybe they might have, but I don't remember. And as I said, I am not a lawyer. I don't pay attention to priority dates. That wasn't my job. I'm a scientific expert, not a priority date expert.

Reply at 3

EX1044, Struhl Dep. 32:19-33:4

Dr. Struhl

Did not know or understand relevance of priority date

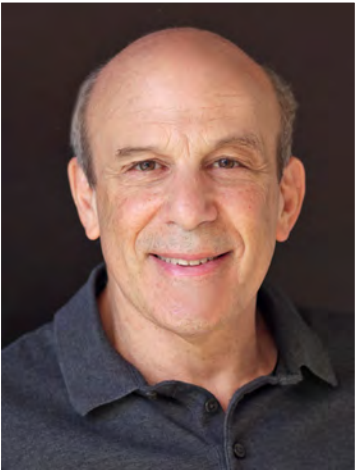


PO's Expert:
Kevin Struhl PhD

- Q. Okay. Now, for the purposes of your opinions in this case, was there any particular time point, you know, any particular historical time point that you used as the baseline time point for when you needed to render your decisions as of? You understand my question?
- A. No.
- Q. You don't understand what I might be getting at?
- A. No.
- Q. Okay. Was there any historical time point that you had to -- that you had to put yourself in, as the time point for rendering your opinions on invalidity?
- A. I was looking at the patent, what the patent said, and what some of the other prior art, particularly patents, said in deciding what the differences were and what was relevant. I wasn't paying attention to years because that was -- again, if you are starting to talk about years that becomes legal. I'm not -- I am not a lawyer. So, you know, if a document was relevant because of the year, I would see it. If it wasn't, I didn't see it. I wasn't making that decision because the fact is, I don't know how to make that decision.

Dr. Struhl

Did not consider the historical development of the art



PO's Expert:
Kevin Struhl PhD

Q. Did you ever do any investigation to figure out when scientists and researchers first become aware of the concept of using multiple colors to identify a single target.

A. As I said, **I didn't do a historical perspective at all.**

Reply at 4

EX1044, Struhl Depo. 58:6-11

Q. ... Do you know when it became known and understood in the field how to detect PCR products using optical means?

A. I don't -- I'm telling you, **I can't give you a year.** When TaqMan probes came out, that was -- that might -- I don't even know if that was the first, but TaqMan probe certainly was that, Okay.

Q. You don't have a historical perspective on when that was?

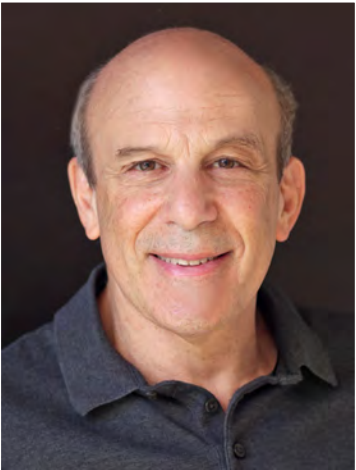
A. No because **why would I care exactly what year that was?**

Reply at 4

EX1044, Struhl Depo. 59:19-60:4

Dr. Struhl

Did not consider the historical development of the art



PO's Expert:
Kevin Struhl PhD

Q. In terms of tuning those probes to have different intensity, when was that technique a technique that was known to the Person of Ordinary Skill in the Art.

A. As I said earlier, I did not do a historical perspective on that.

Reply at 4

EX1044, Struhl Depo. 163:11-16

Q. You didn't investigate that for the purposes of your declaration?

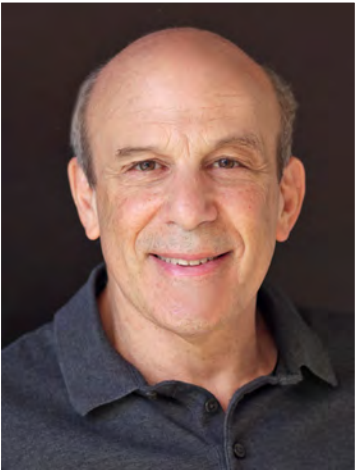
A. I told you, I did not do any historical analysis for anything.

Reply at 4

EX1044, Struhl Depo. 181:6-9

Dr. Struhl

Did not consider the historical development of the art



PO's Expert:
Kevin Struhl PhD

Q. Okay. How about the R0X, CY5 and the CY5.5 dyes, if I were to ask those same questions that I asked earlier about when those dyes became known to scientists and researchers for use with multiplex applications, you wouldn't have any perspective on that; is that right?

A. I have not done historical analysis of when things were first done.

Reply at 4

EX1044, Struhl Depo. 62:15-23 (objections omitted)

Law

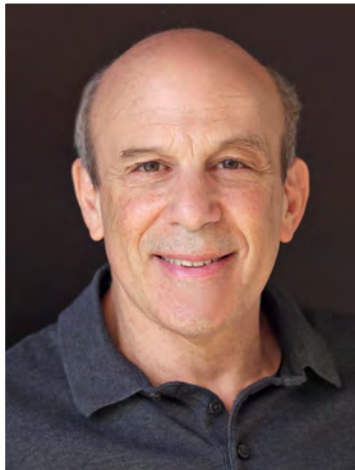
“Critical step” in obviousness analysis is “casting the mind back to the time of invention”



“A **critical step** in analyzing the patentability of claims pursuant to section 103(a) is **casting the mind back to the time of invention**, to consider the thinking of one of ordinary skill in the art, guided only by the prior art references and the then-accepted wisdom in the field.”

In re Kotzab, 217 F.3d 1365, 1369 (Fed. Cir. 2000)

Dr. Struhl



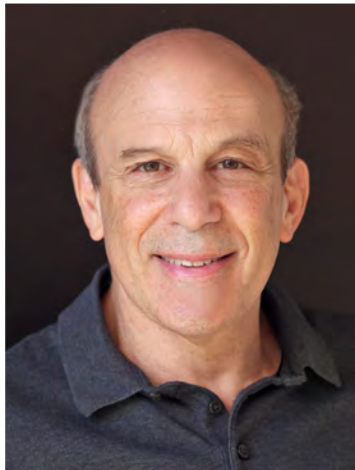
PO's Expert:
Kevin Struhl PhD

Cites the “*Phillips* standard”

21. For purposes of *Inter Partes Review*, I understand that the Board applies the *Phillips* standard, under which claims are to be given their ordinary and customary meaning as understood by a POSA at the time the application was filed. I understand that this analysis is performed by primarily looking to the patent's claims, specification (including drawings), and prosecution history (the “intrinsic” evidence). I understand that if the claim is unclear in view of the intrinsic evidence, then outside (“extrinsic”) evidence also may be considered.

EX2018 Struhl Decl. ¶ 21

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Discusses claim construction

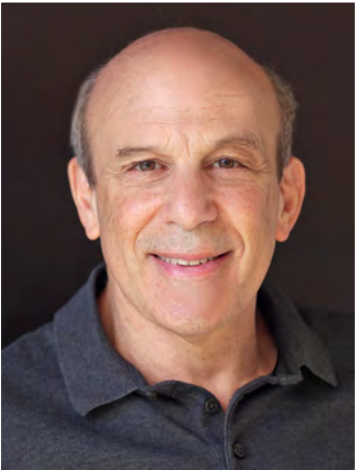
VI. CLAIM CONSTRUCTION

79. For purposes of this declaration, I have reviewed the challenged claims of the '921 Patent in view of their common and ordinary usage and in view of the specification of the '921 Patent.

EX2018 Struhl Decl. ¶ 79

Dr. Struhl

Dr. Struhl did not know the standard for claim construction



PO's Expert:
Kevin Struhl PhD

Q. You don't have any idea why it's - what Phillips is when you refer to Phillips?

A. I assume it's a person.

Reply at 5

EX1044, Struhl Depo. 72:25-73-2

Q. Okay. You don't have opinions on claim construction in this case?

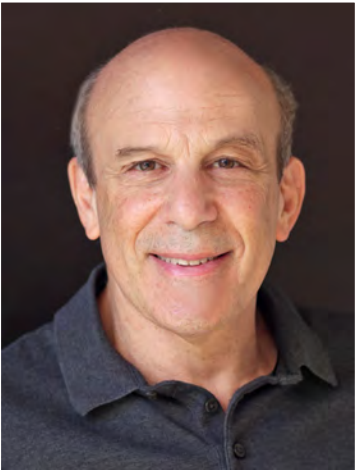
A. No. I haven't thought about it.

Reply at 5

EX1044, Struhl Depo. 83:23-25

Dr. Struhl

Dr. Struhl did not know the standard for claim construction



PO's Expert:
Kevin Struhl PhD

Q. Now, you refer in the second line of this paragraph to something called "Phillips," that's in italics. Do you see that?

A. Yes.

Q. Do you know what Phillips is?

A. No but --

Q. Do you have any idea what you were referring to?

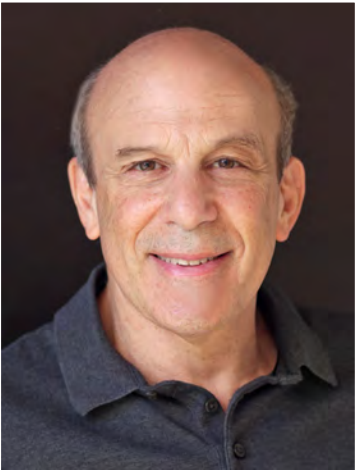
A. This is a legal statement that I was informed of. I don't really know completely what it means. I don't know what the Phillips standard is, other than what it says here.

Reply at 5

EX1044, Struhl Depo. 75:4-15

Dr. Struhl

Dr. Struhl did not know the standard for claim construction



PO's Expert:
Kevin Struhl PhD

- Q. But the only time you remember with any clarity of having heard of the Phillips rule, is when we first began discussing it here today; is that right?
- A. I said I -- in various cases I have done, there are always legal issues that come up such as this, and I don't know the legal issues. I'm not a lawyer. I'm a scientist. It seems that is not as important as the legal stuff in these cases, and I get told stuff.

Reply at 5

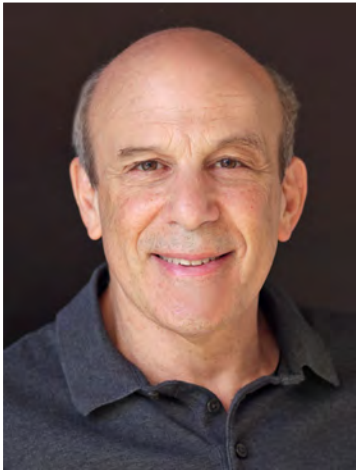
EX1044, Struhl Depo. 74:17-75:3 (objections omitted)

Claims 1-2 of the '921 Patent

1. An assay that is capable of unambiguously detecting the presence or absence of each of at least M analytes, wherein each of said at least M analytes is encoded as a value of one component of a fluorescent signal, thereby generating a coding scheme, wherein said fluorescent signal comprises F wavelength components, and wherein said assay is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in a single sample volume, based on detection of said F wavelength components, when M is greater than F, wherein the assay does not require the use of mass spectrometry or immobilization of said at least M analytes, and wherein the analytes are amplified in the single sample volume.

2. The assay of claim 1, wherein each of said at least M analytes is encoded as at least one first value in a first component of said fluorescent signal and at least one second value in a second component of said fluorescent signal.

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

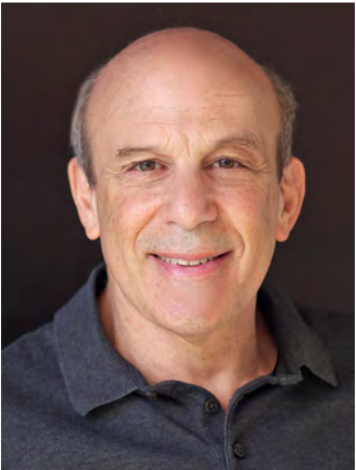
Dr. Struhl did not understand key claim elements

- Q. Well, it says -- it says, "each of said at least M analyte is encoded as a value of one component of a fluorescent signal," it says. So each of the analyte is encoded as one component of a fluorescent signal. How might you do that?
- A. I mean, the analyte is encoded by the components that are the light and the intensity.
- Q. Well, this says, "one component." What do you understand it to be referring to when it says, "one component"? Do you know?
- A. I must say I don't really completely understand what this -- this particular sentence means. I understand what the patent is doing. I understand the basic idea. I'm not sure what they meant by "one component," because there are typically the coding scheme for -- Each analyte has a coding scheme, which has got to do with the colors and the intensity, and that's clear. So I'm not sure what their -- the way this is written, I'm not sure what they meant.

Reply at 5

EX1044, Struhl Depo. 97:16-98:11

Dr. Struhl



PO's Expert:
Kevin Struhl PhD

Dr. Struhl did not understand key claim elements

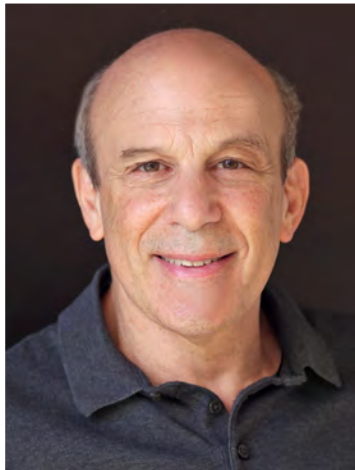
"I don't think the patent is really concerned about number of components. It's concerned what the final answer is."

Reply at 5

EX1044, Struhl Depo. 96:12-14

Dr. Struhl

Repeatedly refers to claim 2



PO's Expert:
Kevin Struhl PhD

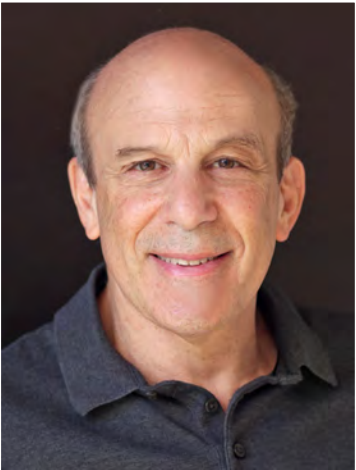
B. Ground 2 (Claims 2, 4, 6-7, and 14-19): Obviousness in View of
Larson

96. The Petition identifies dependent claims 2, 4, 6-7, and 14-19 as allegedly being obvious over Larson. Petition at 44-51. However, my analysis and views set forth above in ¶¶83-95 of this Declaration regarding Ground 1, describing the many material differences between Larson and claim 1, apply equally (if not more) to claims 2, 4, and 6-7 which depend from claim 1, to independent claim 14 which recites similar limitations as discussed above for claim 1, and to claims 15-19 which depend from claim 14.

EX2018 Struhl Decl. ¶ 96

Dr. Struhl

Did not understand claim 2



PO's Expert:
Kevin Struhl PhD

Q. Okay. So you are aware that this claim, Claim 2, is not limited to encoding each -- this is -- this is -- this **Claim 2** is not limited to encoding each analyte with one component, fair?

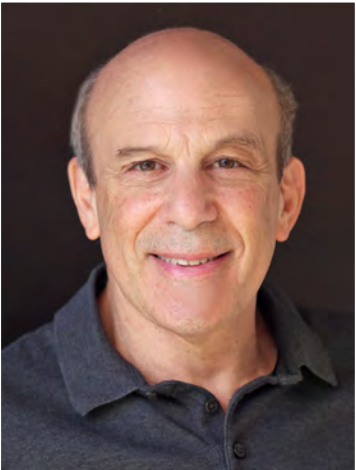
A. Yeah. I don't know what you are getting at. I -- in fact, **I probably shouldn't even be opining on Claim 2 since I never looked at it before.** I looked at it, but I didn't opine on it.

Reply at 5

EX1044, Struhl Depo. 100:14-23 (objections omitted)

Dr. Struhl

Did not understand claim 2



PO's Expert:
Kevin Struhl PhD

- Q. Is it accurate that Claim 2 broadens out Claim 1 to encompass not just one component but two components?
- A. I don't think it -- I mean, I don't think it broadens it; if anything, it's restricting it. That's why it's Claim 2 and not Claim 1.
- Q. Okay. How does it restrict things relative to Claim 2?
- A. I'm not going to get into the difference between Claim 1 and 2. I haven't analyzed Claim 2.

Reply at 5

EX1044, Struhl Depo. 101:8-21 (objections omitted)

Petitioner's Expert

Repeatedly cites PO's infringement contentions and coding allegations



Petitioner's Expert:
Carl Batt PhD

71. The accused assays and kits include simple assays that detect three target sequences using only two fluorophores to generate different color signals. *See, e.g., id.* at 10 (“In the SARS-CoV-2 Kit, N1 is encoded as FAM positive, N2 as FAM and HEX positive, and RP as HEX positive.”), 11 (“In the SARS-CoV-2 Wastewater Quantification Kit, N2 is encoded as FAM positive, E as FAM and HEX positive, and NHV as HEX positive.”), 14, 21, 25, 30-33, 38, 50, 52-53. For example, Patent Owner contends, consistently throughout its infringement contentions, that coding schemes fitting this description and as shown below satisfy the limitations of the '921 patent to constitute infringement:

Analyte	FAM	HEX
N1	1	0
N2	1	1
RP	0	1

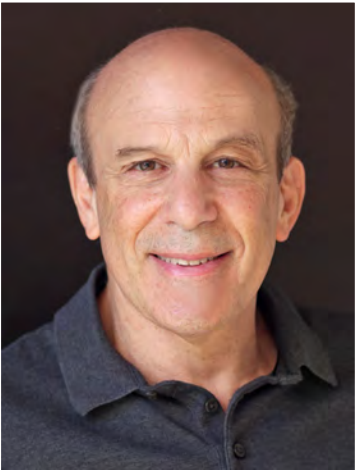
id. at 10 (depicting coding scheme in SARS-CoV-2 Kit);

Analyte	FAM	HEX
N2	1	0
E	1	1
MHV	0	1

id. at 11 (depicting coding scheme in SARS-CoV-2 Wastewater Quantification Kit (“Wastewater Kit”).

Dr. Struhl

Did not understand relevance of infringement contentions



PO's Expert:
Kevin Struhl PhD

Q. Okay. Then why did you review the infringement contentions for your case?

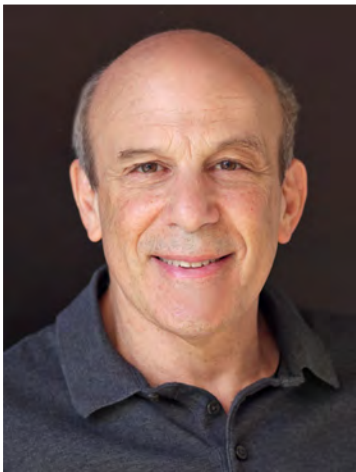
A. I actually don't know the answer. Maybe just to know that there was an infringement case. I don't really know why. You know, I don't produce the documents on my own. So, you know, if I was going to have an opinion about an infringement, I would be – I would have a major opinion about infringement. As you can see, I don't have anything about infringement in my declaration.

Reply at 6

EX1044, Struhl Depo. 111:16-25

Dr. Struhl

Did not understand relevance of infringement contentions



PO's Expert:
Kevin Struhl PhD

- Q. Okay. Page 7, the original page numbering in the middle of the page, you state that you reviewed Exhibit 1039, Exhibit 1040. Those are infringement claim charts from infringement contentions. Do you see that?
- A. I do.
- Q. Those are documents you reviewed as well?
- A. Very cursory. I said I didn't spend almost any time on infringement.
- Q. Do you know if those are Caltech's or Bio-Rad's infringement for drugs?
- A. Again, I said I didn't spend almost any time on infringement. I really don't know who is doing what about --
- Q. Why did you review those infringement claim charts?
- A. I don't remember, actually.
- Q. Okay.
- A. I'm not sure I reviewed that. I mean, you know, I saw the documents. I spent almost no time on infringement. I have no opinions on infringement. So I really don't remember anything about it.

BIO-RAD

Reply at 6

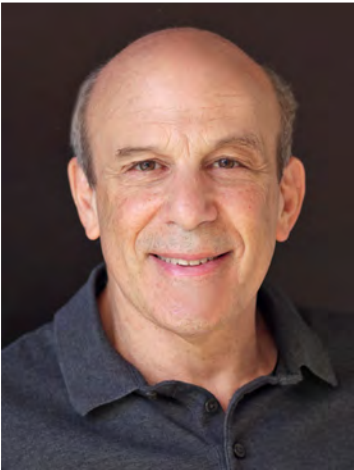
EX1044, Struhl Depo. 106:2-24

DEMONSTRATIVE EXHIBIT – NOT EVIDENCE

131

Dr. Struhl

Did not understand relevance of infringement contentions



PO's Expert:
Kevin Struhl PhD

- Q. ... When you reviewed them, when you reviewed these infringement contentions, did you identify anything that you thought was erroneous or needed to be changed?
- A. I reviewed these so cursorily. This was not really -- it's not really my thing. We are not doing -- I was not involved in the infringement things, whatever they are. So I didn't pay much attention to this. I certainly didn't read it cover to cover in any detail. It wasn't any point to doing that.

Reply at 6

EX1044, Struhl Depo. 109:1-11

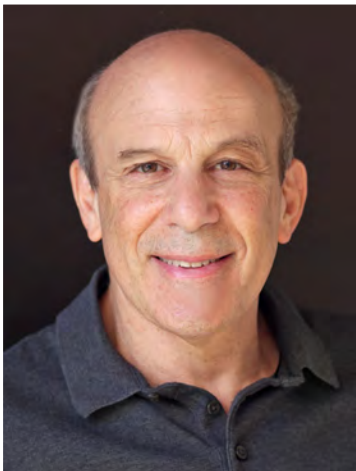
- Q. Do you have any idea as to how many components of the fluorescent signal are used to encode that analyte?
- A. I told you I'm not giving you an opinion on this. I didn't review it. I didn't have any comments about infringement. I don't know what these kits are. I am not going to give you an opinion without -- you know, this would take a long time to give a real opinion. Infringement cases are serious business. I had no part of it.

Reply at 6

EX1044, Struhl Depo. 111:3-15 (objections omitted)

Dr. Struhl

Did not understand relevance of infringement contentions

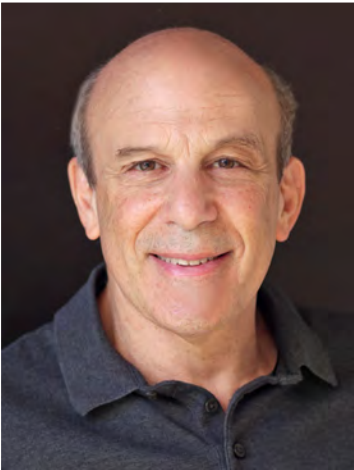


PO's Expert:
Kevin Struhl PhD

- Q. Did you review Dr. Batt's declaration in this case?
- A. The 1002, yes.
- Q. Okay. Do you know if he said anything about the infringement contentions?
- A. I have to look. Maybe -- I don't know if he did or he didn't.
- Q. Okay. But one thing for sure, If he said something about the infringement contentions, you don't have a responsive opinion on that because you hardly spent any time looking at those, right?
- A. I told you, I didn't spend any time on infringement. That's a whole separate case.
- Q. Okay?
- A. Maybe multiple cases, for all I know, and so I didn't spend any time on it. So anything said about that, he -- I just didn't pay attention to it because I didn't comment on it.
- Q. Okay. And the same thing was true to the extent the Petitioner relied upon Caltech's infringement contentions in this case, you paid no attention to that?
- A. My role in this case was a scientific evaluation of what the patent was, and in consideration of other patents that were, you know, considered relevant. That's it. I had nothing to do with infringement.
- Q. Okay. To the extent Dr. Batt rendered opinions based on Patent Owner's infringement contentions, did you present any rebuttal?
- A. I don't remember discussing anything about infringement in my responses.

Dr. Struhl

Did not understand relevance of infringement contentions



PO's Expert:
Kevin Struhl PhD

Q. You see there, there is an encoding scheme for the "SARS-CoV-2 Kit"?

A. I have not seen -- I have not paid attention to this before. I'm not even sure I have seen it.

Q. Okay. Go ahead and look at it. Do you see that encoding scheme there for the three different analytes?

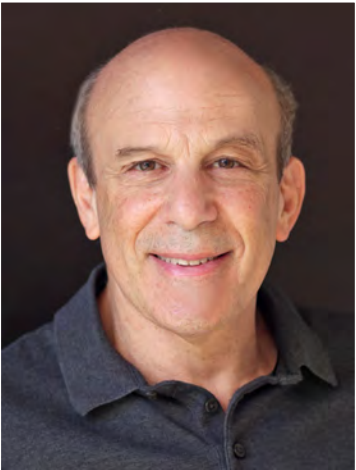
A. Yeah. I'm not willing to comment on this. I don't know what this -- this is out of context.

Q. Well, let me ask you this, you reviewed this for your declaration, right?

A. It's not in my declaration.

Dr. Struhl

Did not understand relevance of infringement contentions



PO's Expert:
Kevin Struhl PhD

Q. Let's go back to the invalidity infringement contentions, Exhibit 1012. Let's take a look at Page 30.

A. I'm on Page 30.

Q. You see there is a table there for the SARS-Co -- COVID 2 wastewater quantification kit?

A. I see there is a table.

Q. Have you ever seen that table before?

A. I don't remember.

Reply at 6

EX1044, Struhl Depo. 114:23-115:6

Law

Reliance on contradictory and unreliable testimony is an abuse of discretion



“The Board **abused its discretion** by improperly crediting expert testimony that has **multiple material contradictions** and that the Board found **lacked credibility** at least in certain respects.”

Interactive Comms. Intl., Inc. v. Blackhawk Network, Inc., IPR2024-00465, Paper 39 at 3 (Director Oct. 1, 2025))

PO's Motion To Strike

The Petition

Was clear that Larson teaches multiple targets in a single droplet

Indeed, Larson expressly teaches that there may be multiple targets in a single individual droplet and that its methods accommodate such “multiple occupancy” situations. See EX1017 ¶¶ 129, 141; EX1002 ¶ 321. Larson’s assays use conditions and parameters that do precisely this. EX1002 ¶ 321. While this alone proves obviousness, the contemporaneous Saxonov reference likewise teaches such “multiple occupancy” situations and that one can easily distinguish among droplets having different combinations of targets based on their intensity in different color channels. Dr. Batt confirms a POSA would have at least found it obvious to incorporate Saxonov’s teachings into Larson to do this. EX1002 ¶¶ 318-352.

Petition at 52

The Petition

Was clear that Larson teaches multiple targets in a single droplet

While Larson focuses on scenarios where droplets have only one target, it also teaches scenarios where individual droplets have multiple targets. Specifically, Larson teaches that the number of targets present in droplets will follow a Poisson distribution. EX1017 ¶ 6; EX 1002 ¶ 331. As Larson explains, in a so-called “limiting dilution” regime, “some reactions contain zero DNA molecules, some reactions contain one molecule, and *frequently some other reactions contain multiple molecules*, following the Poisson distribution.” EX1017 ¶ 6. Larson goes

Petition at 54-55

The Petition

Was clear that Larson teaches multiple targets in a single droplet

multiple molecules, following the Poisson distribution.” EX1017 ¶ 6. Larson goes on to state that “the invention is compatible with all distributions of DNA loading that conform to limiting or terminal dilution.” *Id.* ¶ 52. Thus, by explaining that the invention is compatible with “limiting” dilution, Larson makes clear that the invention works in situations where droplets may contain multiple targets. Indeed, Larson later refers to the possibility of such “multiple occupancy” situations, and, while noting that complications may arise, says that “methods of the invention can accommodate these complications arising from multiple occupancy.” *Id.* ¶ 141; EX1002 ¶ 332.

Petition at 52

Patent Owner's Response

PO contends Larson is limited to single occupancy droplets

a. **Larson Teaches Eliminating the Very “Multiple Occupancy” Situations to Which Petitioner Points.**

Petitioner's Ground 3 alleges Larson teaches “there may be multiple targets in a single individual droplet and that its methods accommodate such ‘multiple occupancy’ situations,” stating “Larson’s assays use conditions and parameters that do precisely this.” Petition, 52, *citing* EX1017, ¶¶129, 141; EX1002, ¶321.

However, Larson’s actual disclosure about accommodating “‘multiple occupancy’ situations” is *incompatible* with an assay that is capable of unambiguously detecting the presence or absence of each of said at least M analytes, in any combination of presence or absence, in the single sample volume in which the analytes are amplified. EX2018, ¶99; *see* EX1001, 67:16-20, 68:31-34.

Law

Elaboration and new evidence are not prohibited



“Parties are **not barred** from elaborating on their arguments on issues previously raised.”

Chamberlain Grp., Inc. v. One World Techs., Inc., 944 F.3d 919, 925 (Fed. Cir. 2019)

Law

Elaboration and new evidence are not prohibited



“The Board's **error** was **parsing** Ericsson's arguments on reply with **too fine of a filter.**”



“Ericsson does not identify a previously unidentified piece of prior art to make a meaningfully distinct contention, but instead **expands the same argument made in its Petition.**”

Ericsson Inc. v. Intell. Ventures I LLC, 901 F.3d 1374, 1380-81(Fed. Cir. 2018)

Law

Elaboration and new evidence are not prohibited



“Since the need to rely on new evidence in response may not arise until a particular point has been raised in the patent owner response or the institution decision, we have held that there is no blanket prohibition against the introduction of new evidence during an IPR.”

Rembrandt Diagnostics, LP v. Alere, Inc., 76 F.4th 1376, 1384 (Fed. Cir. 2023)

Law

Elaboration and new evidence are not prohibited



“There is **no requirement...to anticipate and set forth every legal or factual issue** that might arise in the course of the trial.”



“However, **the introduction of new evidence in the course of the trial is to be expected** in inter partes review trial proceedings and, as long as the opposing party is given notice of the evidence and an opportunity to respond to it, **the introduction of such evidence is perfectly permissible** under the APA.”



“The **development of evidence** in the course of the trial is in keeping with the oppositional nature of an inter partes review proceeding.”

Law

Elaboration and new evidence are not prohibited



“Apple’s legal ground did not change in its reply—its reply still asserted that claims 6–9 would have been obvious over Hirsch and Martin.”

Apple Inc. v. Andrea Elecs. Corp., 949 F.3d 697, 706 (Fed. Cir. 2020)

Thank You