

# 10X Genomics Inc. (Petitioner) v. Bio-Rad Laboratories, Inc. (Patent Owner)

IPR2020-00087 (U.S. Patent No. 9,562,837) and  
IPR2020-00089 (U.S. Patent No. 9,896,722)

## Petitioner's Demonstratives Thorsen Grounds



January 27, 2021

# Thorsen

## Microfluidic Technologies for High-Throughput Screening Applications

Thesis by  
Todd Thorsen

In Partial Fulfillment of the Requirements  
For the Degree of  
Doctor in Philosophy

California Institute of Technology  
Pasadena, California

2003

(Defended September 23, 2002 )

## Microfluidic Technologies for High-Throughput Screening Applications by Todd Thorsen, 2002 (“Thorsen”)

- Discloses droplet-based microfluidic devices for high-throughput chemical screening
- Teaches both an “incubation chamber” and “outlet wells” – each of which independently discloses the claimed “separation chamber” under Patent Owner’s constructions

IPR2020-00087 Ex. 1003 (Thorsen), Petition at 12-15  
IPR2020-00089 Ex. 1003 (Thorsen), Petition at 13-15

# Thorsen-Based Grounds

**All Thorsen-based Grounds apply Patent Owner's claim constructions**

Grounds	References	Claims
1a	Thorsen, single operation (§ 102)	837 Patent: 1-4, 10-13, 19 722 Patent: 1-3, 10-11, 14
1b	Thorsen, Chapter 3 system (§ 102)	837 Patent: 1-4, 10-13, 19 722 Patent: 1-3, 10-11, 14
1c	Thorsen, Chapter 4 system (§ 102)	837 Patent: 1-2, 4, 10-13 722 Patent: 1-3, 10-11, 14
2	Thorsen (§ 103)	837 Patent: 1-4, 7-13, 19 722 Patent: 1-4, 10-11, 14
3	Thorsen + Ismagilov 119 (§ 103)	837 Patent: 1-2, 4, 7-13, 19 722 Patent: 1-7, 10-17
4	Thorsen + Ismagilov 091 + Ismagilov 119 (§ 103)	837 Patent: 1-2, 4, 7-13, 19 722 Patent: 1-7, 10-17

IPR2020-00087 Petition at 19-27  
IPR2020-00089 Petition at 20-27

# 837 Patent – Independent Claim 1

1. An assembly, the assembly comprising:

**[1.1]** a microchannel in a horizontal plane;

**[1.2]** at least one droplet formation module comprising a sample inlet, an immiscible fluid inlet, and a junction, wherein the junction is located between the sample inlet and the microchannel and the droplet formation module is configured to produce droplets comprising the sample surrounded by the immiscible fluid; and

**[1.3]** at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet,

**[1.4]** wherein the separation chamber is upright to the microchannel and out of the horizontal plane;

**[1.5]** wherein the droplet formation module and the separation chamber are in fluid communication with each other via the microchannel; and

**[1.6]** wherein the separation chamber has a wider cross-section than the microchannel cross-section to accumulate a plurality of droplets comprising the sample and

**[1.7]** is of a volume sufficient to separate the plurality of droplets comprising the sample from the immiscible fluid within the separation chamber.



# 722 Patent – Independent Claim 1

1. A method of reducing contamination associated with sample handling, comprising:
  - [1.1]** providing an aqueous fluid comprising a sample through a sample inlet;
  - [1.2]** providing an immiscible fluid flowing through a main channel that is in fluidic communication with the sample inlet, wherein the main channel is in a horizontal plane;
  - [1.3]** partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a sample;
  - [1.4]** flowing the droplets toward a downstream separation chamber that is in fluidic communication with the main channel,
  - [1.5]** wherein the separation chamber has a wider cross-section than the main channel cross-section and
  - [1.6]** the separation chamber is disposed perpendicular to the main channel; and
  - [1.7]** separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.

# 722 Patent – Independent Claim 14

14. A method of reducing sample loss associated with sample handling, comprising:
- [14.1]** providing an aqueous fluid comprising a sample through a sample inlet;
  - [14.2]** providing an immiscible fluid flowing through a main channel that is in fluidic communication with the sample inlet, wherein the main channel is in a horizontal plane;
  - [14.3]** partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a sample;
  - [14.4]** flowing the droplets toward a downstream separation chamber that is in fluidic communication with the main channel,
  - [14.5]** wherein the separation chamber has a wider cross-section than the main channel cross-section and
  - [14.6]** the separation chamber is disposed perpendicular to the main channel; and
  - [14.7]** separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.

# Claim Constructions

Term or Phrase	Petition's Constructions (based on Bio-Rad's Infringement Contentions)	Bio-Rad's Proposed Construction
<b>"separation chamber"</b> (837 Patent, Claim 1; 722 Patent, Claims 1, 14)	Broad enough to encompass an open well	"chamber sized to accommodate multiple droplets and an immiscible fluid in separated form"
<b>"sample"</b> (837 Patent, Claim 1; 722 Patent, Claims 1, 14)	"a compound, composition, and/or mixture of interest, from any suitable source(s)"	"fluid containing molecules, cells, small molecules (organic or inorganic) or particles"
<b>"droplet receiving outlet"</b> (837 Patent, Claim 1)	Broad enough to include the opening at the top of an open outlet well	"portion of the separation chamber where droplets are collected"

IPR2020-00087 Petition at 17-19, POR at 16-25  
IPR2020-00089 Petition at 18-19, POR at 15-22

# Claim Constructions

Term or Phrase	Petition's Constructions (based on Bio-Rad's Infringement Contentions)	Bio-Rad's Proposed Construction
<b>"coupled"</b> (837 Patent, Claims 7-8)	Broad enough to encompass transporting by pipette between an alleged outlet/chamber and vessel	<b>"operably linked"</b>
<b>"polymerase chain reaction"</b> (837 Patent, Claim 13; 722 Patent, Claim 7)	Broad enough to encompass any amplification reaction	<b>"methods by K.B. Mullis for increasing concentration of a segment of a target sequence in a mixture of genomic DNA without cloning or purification"</b>

IPR2020-00087 Petition at 17-19, POR at 16-25  
 IPR2020-00089 Petition at 18-19, POR at 15-22



# Incubation Chamber: 3 Disputes (independent claims)

Patent Owner disputes only three limitations as to Thorsen's Incubation Chamber for the independent claims:

- (1) "separation chamber" (837 Patent & 722 Patent)
- (2) "droplet receiving outlet" (837 Patent)
- (3) "sample" (837 Patent & 722 Patent) (not argued in sur-reply)

IPR2020-00087 POR at 25-31, 37-41, Pet. Reply at 5, PO Sur-Reply at 4-7  
IPR2020-00089 POR at 23-27, Pet. Reply at 4, PO Sur-Reply at 4-7

# Incubation Chamber: 3 Disputes (independent claims)

- (1) “separation chamber” (837 Patent & 722 Patent)
  - No dispute that Thorsen’s incubation chamber meets the size and orientation requirements of the claims
  - Bio-Rad argues only that Thorsen’s incubation chamber does not separate droplets from oil
  - Bio-Rad’s construction of “separation chamber” is “chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”
- (2) “droplet receiving outlet” (837 Patent)
- (3) “sample” (837 Patent & 722 Patent) (not argued in sur-reply)

IPR2020-00087 POR at 25-31, 37-41, Pet. Reply at 7-12, Petition at 45-59  
IPR2020-00089 POR at 23-27, Pet. Reply at 6-10, Petition at 44-57

# Incubation Chamber: “separation chamber”

## **PO’s construction:**

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.7]:**

“...is of a volume sufficient to separate the plurality of droplets comprising the sample from the immiscible fluid within the separation chamber.”

## **722 Patent [1.7]/[14.7]:**

“separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.”

mineral oil and its elastomeric properties. A significant design modification was added to the device to accommodate chemical screening reactions where incubations of several minutes to a few hours are optimal (Figure 3.28A). The serpentine output channel at the crossflow junction was replaced with a large ( $\sim 500\ \mu\text{m}$ ) high cavity to slow down the droplets and create an on-chip incubation chamber (Figure 3.28B). This design element not only created a more flexible on-chip screening environment, but also significantly reduced the footprint of the chip from  $1\ \text{in}^2$  to  $0.5\ \text{in}^2$ . A control layer of valves was added for sorting droplets at the T-junction intersection at the end of the chip (Figure 3.28C). The devices were fabricated from the rounded 5740 photoresist molds. The incubation cavity was created by placing  $\sim 1\ \mu\text{l}$  of 5740 resist over the center of the developed, but not hard baked, flow layer mold. The mold was then hard baked for 30 minutes at  $120^\circ\text{C}$ .

IPR2020-00087 Ex. 1003 (Thorsen) at 99, 101, Petition at 45-47, 57-58  
IPR2020-00089 Ex. 1003 (Thorsen) at 99, 101, Petition at 44-46, 55-56

# Incubation Chamber: “separation chamber”

## **PO’s construction:**

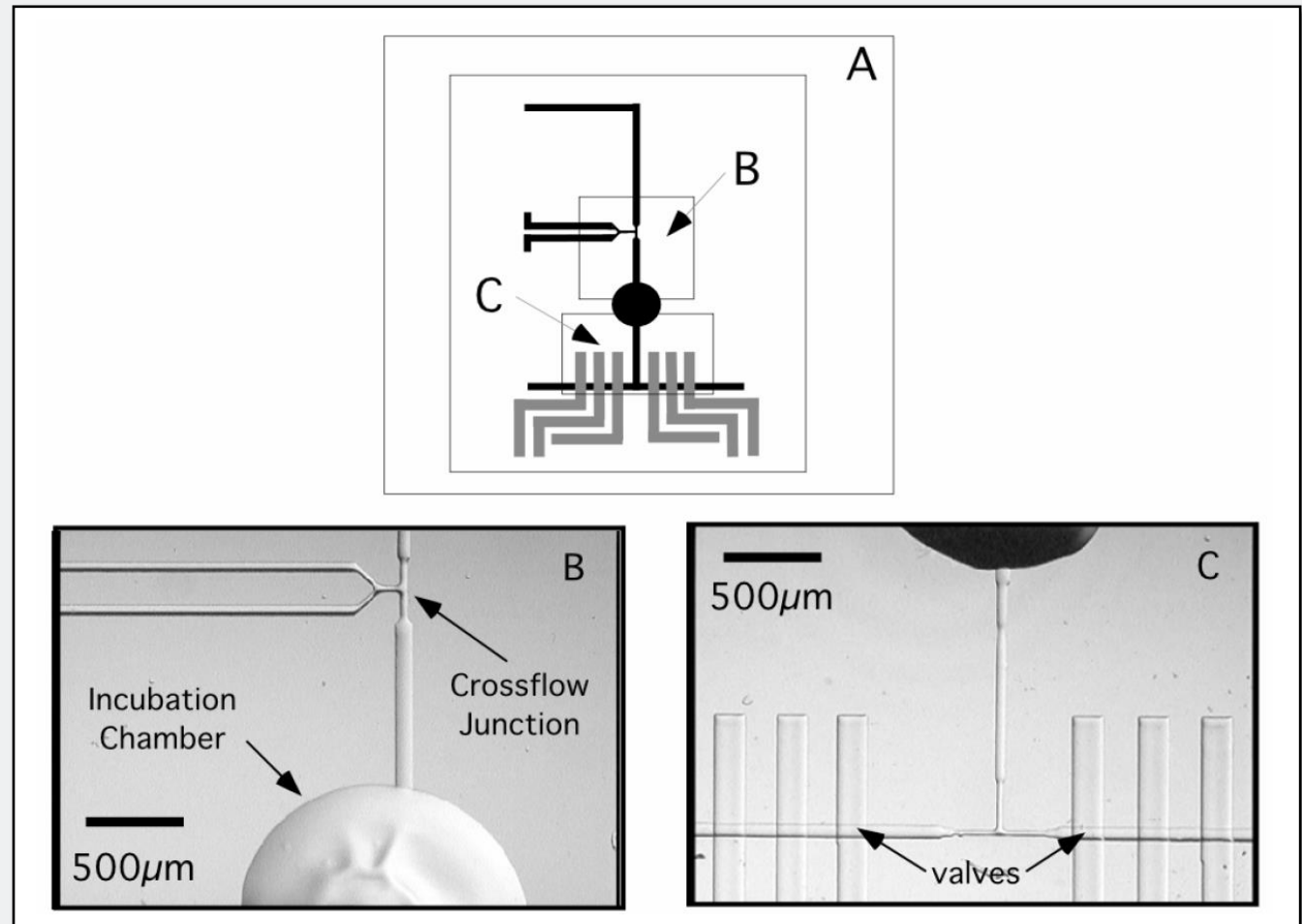
“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.7]:**

“...is of a volume sufficient to separate the plurality of droplets comprising the sample from the immiscible fluid within the separation chamber.”

## **722 Patent [1.7]/[14.7]:**

“separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.”



IPR2020-00087 Ex. 1003 (Thorsen) at 101 Fig. 3.28, Petition at 45-47, 57-58  
IPR2020-00089 Ex. 1003 (Thorsen) at 101 Fig. 3.28, Petition at 44-46, 55-56



# Incubation Chamber: “separation chamber”

## **PO’s construction:**

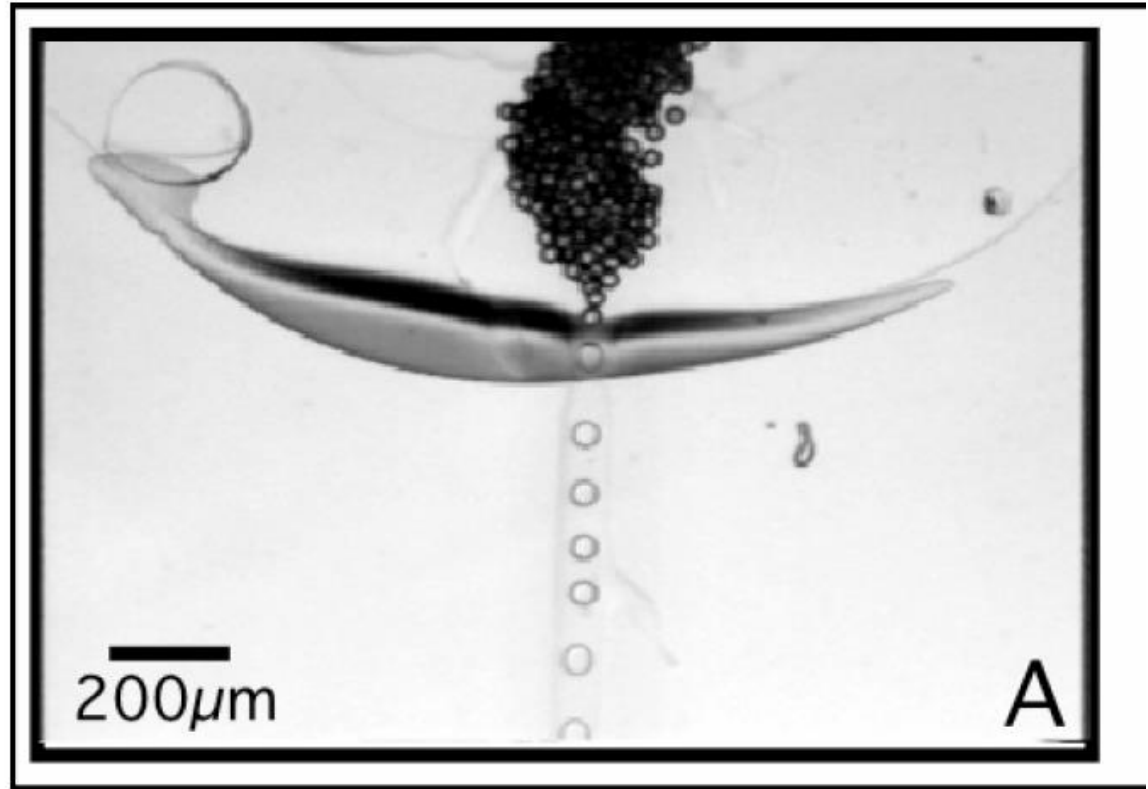
“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.7]:**

“...is of a volume sufficient to separate the plurality of droplets comprising the sample from the immiscible fluid within the separation chamber.”

## **722 Patent [1.7]/[14.7]:**

“separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.”



**Figure 3.29:** A) Water droplets entering incubation cavity of Sylgard 184 device.

B) Sorting of droplets using elastomeric valves.

IPR2020-00087 Ex. 1003 (Thorsen) at 103 Fig. 3.29, Petition at 45-47, 57-58

IPR2020-00089 Ex. 1003 (Thorsen) at 103 Fig. 3.29, Petition at 44-46, 55-56

# Incubation Chamber: “separation chamber”



**Dr. Richard Fair**



“Thorsen discloses an incubation chamber of sufficient volume that the droplets can move relative to the oil phase. *Id.*, Figs. 3.28, 3.29, 99-103. Because the droplets and oil are of different densities, the Thorsen droplets will sink relative to the less dense mineral oil—indeed, Thorsen shows this in Figure 3.29 (showing sinking droplets accumulating on the floor of the incubation chamber while the oil floats above). See section VI (“State of the Art”) (discussing the well-known densities of water, fluorinated oil, and mineral oil); Ex. 1003, Figs. 3.28, 3.29, 99-103. Thorsen discloses that the incubation chamber has a volume of  $\sim 1 \mu\text{L}$  while the droplets have a volume of  $\sim 30 \text{ pL}$  ... which a person of ordinary skill in the art would know to be (and to disclose) a volume sufficient to allow the droplets to separate under Bio-Rad’s interpretation. A person of ordinary skill in the art would know that there are 1,000,000 picoliters (pLs) in a single microliter ( $\mu\text{L}$ ) so Thorsen’s incubation chamber has a volume sufficient to accommodate as many as  $\sim 300,000$  droplets...”

IPR2020-00087 Ex. 1008 at ¶ 154  
see also IPR2020-00089 Ex. 1008 at ¶ 157 (similar)

# Incubation Chamber: “separation chamber”



**Dr. Richard Fair**



“It was also well-known to generate droplets using fluids of different densities... Aqueous (water) solutions and fluorinated oils have different densities... Water has a density of 1 g/cm<sup>3</sup>, while perfluorohexane and perfluorodecaline have densities of around 1.7 and 1.9 g/cm<sup>3</sup>, respectively. Ex. 1014, 4-5. These are constant, unchanging properties of these chemicals, and this difference in density means that the less dense water droplets necessarily will float to the top in denser oil. By contrast, mineral oil is less dense than water (between 0.8 and 0.9 g/ml), and so aqueous droplets would sink to the bottom. Ex. 1015, Ex. 1016, Ex. 1063, Ex. 1064.”

IPR2020-00087 Ex. 1008 at ¶ 51 (Section VI. State of the Art)

IPR2020-00089 Ex. 1008 at ¶ 51 (Section VI. State of the Art)

# Incubation Chamber: “separation chamber” Institution Decision

**Institution Decisions found that the incubation chamber discloses the claimed “separation chamber”:**

With this understanding of Thorsen’s experimental setup, Patent Owner’s assertion that Thorsen Figure 3.29 shows the droplets “in focus” and “in the same plane” in the main channel and incubation chamber (Prelim. Resp. 28) appears to support Petitioner’s position that the figure shows “sinking droplets accumulating on the floor of the chamber while the oil floats” (Pet. 57). Patent Owner’s argument is consistent with the droplets having sunk to the bottom in both the incubation chamber and the microchannels, with the droplets resting on the coated coverslip and the oil floating above the droplets. *Because the microscope is inverted in Thorsen’s*

IPR2020-00087 Paper 9 (Institution Decision) at 35-37  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 29-33 (similar)



# Incubation Chamber: “separation chamber” Institution Decision

## Institution Decisions found that the incubation chamber discloses the claimed “separation chamber”:

Third, Patent Owner’s argument that the droplets and oil “do not separate” in Thorsen’s incubation chamber (Prelim. Resp. 30) is belied by its earlier arguments that Figure 3.29 shows the droplets “clumped together” and “bunch[ed] together in the incubation chamber” (*id.* at 28, 29).

Moreover, Patent Owner does not dispute Petitioner’s premise that, because Thorsen’s droplets and mineral oil have different densities, the droplets will sink relative to the less dense mineral oil. Pet. 57.

For all of these reasons, and for purposes of institution, we are persuaded that Petitioner has sufficiently shown that Thorsen Chapter 3 discloses the “separation chamber” recited in claim 1.

IPR2020-00087 Paper 9 (Institution Decision) at 35-37  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 29-33 (similar)

# Incubation Chamber: “separation chamber” Bio-Rad’s Argument

PO’s Sur-Reply argument that 100% separation of droplets and oil is required contradicts PO’s prior arguments to the District Court and this Board:

29-31). Patent Owner explained that there can be no separation when oil remains mixed in between the droplets while in the incubation chamber. (*See id.*). Patent Owner further explained that there can be no separation when the same amount of droplets and oil enter and leave the chamber *together*. (*See id.*). In sum, the incubation chamber of Thorsen Chapter 3 cannot be considered a “separation chamber” when the material leaving the incubation chamber is a *mixture* of droplets and oil, not a *separation* of droplets and oil. (*See id.*). Again, Petitioner does not

IPR2020-00087 PO Sur-Reply at 6  
IPR2020-00089 PO Sur-Reply at 6

# Incubation Chamber: “separation chamber” Bio-Rad’s Inconsistent District Court Position

In District Court, PO argued for the same claim construction of “separation chamber” for the 837 and 722 Patents that PO advances here:

## **Bio-Rad's Proposed Construction**

chamber sized to accommodate multiple droplets and an immiscible fluid in separated form

IPR2020-00087 Ex. 1082 at 5, Pet. Reply at 8

# Incubation Chamber: “separation chamber” Bio-Rad’s Inconsistent District Court Position

In District Court, PO disclaimed that separation must occur for something to be a “separation chamber”:

**“The claims of the ‘837 Patent are directed to an assembly. They do not require separation to occur.”**

IPR2020-00087 Ex. 1082 (Bio-Rad’s District Court Claim Construction Reply) at 12, Pet. Reply at 8



# Incubation Chamber: “separation chamber”

## Bio-Rad’s Inconsistent Argument To This Board

To this Board, Bio-Rad argued **against** a claim construction requiring “substantially only droplets” to exit the separation chamber:

Term or Phrase	Bio-Rad’s Proposed Construction	10X’s Proposed Construction in IPR2020-00086
<b>“droplet receiving outlet”</b> (837 Patent, Claim 1)	“portion of the separation chamber where droplets are collected”	<i>Subject to 112(f):</i> <i>Structure:</i> the outlet structure described at 3:46-58, 53:21-35, or outlet 805 as described at 54:34-46, and Figure 38 and equivalents thereof  <i>Function:</i> receiving substantially only droplets when exiting from the chamber  <i>Alternative:</i> “a structure connected to the interior space of the chamber and configured so that substantially only droplets flow into it when exiting the chamber”

IPR2020-00086 Paper 26 (POR) at 25, Paper 2 (Petition) at 18-19;  
IPR2020-00087 Paper 30 (Pet. Reply) at 8, 13

# Incubation Chamber: “separation chamber” Bio-Rad’s Inconsistent Argument To This Board

To this Board, Bio-Rad argued **against** a claim construction requiring “substantially only droplets” to exit the separation chamber:

*Finally*, the Board should affirm its rejection of Petitioner’s multiple alternative constructions for “droplet receiving outlet.” (*See* Pet. At 19, 21-22). The Board correctly held that that “droplet receiving outlet” conveys sufficiently definite structure in which the droplets are collected inside the separation chamber, not the function of retrieving (or “receiving”) the droplets from the separation chamber at Petitioner contended. (Dec. at 13-14; Ex. 2016 at ¶¶105-108). Furthermore, the Board correctly rejected Petitioner’s second alternative construction requiring that “substantially only droplets” exit the chamber through the outlet because it is inconsistent with the specification and dependent claim 10 of the ‘837 patent. (*Id.* at 14-15).

IPR2020-00086 Paper 26 (POR) at 25;  
IPR2020-00087 Paper 30 (Pet. Reply) at 8, 13

# Incubation Chamber: “separation chamber” Bio-Rad’s Inconsistent Argument To This Board

To this Board, Bio-Rad argued **against** a claim construction requiring “substantially only droplets” to exit the separation chamber:

fluid.”); Ex. 2016 at ¶¶97-100). The ‘837 specification also discusses specific locations of the “droplet receiving outlet” within the chamber. For example, the [ . . . ]

When the droplets are more dense than the immiscible fluid, they fall, and the “droplet receiving outlet” may be located at the bottom of the chamber. (*Id.* at 4:12-16 (“In another example, aqueous droplets placed in a chamber of certain mineral oils will tend to sink due to their greater density. Accordingly, an outlet for collecting the displaced droplets can be positioned at the bottom of the chamber.”))). The ‘837 specification also explains that the “droplet receiving outlet” may be placed in between the top and bottom of the chamber. (*Id.* at 3:51-58 (“The outlet may be at the top, middle, or bottom of the chamber . . . .”))).

IPR2020-00086 Paper 26 (POR) at 22-23;  
IPR2020-00087 Paper 30 (Pet. Reply) at 8, 13

# Incubation Chamber: "separation chamber"

## The Specification Contradicts Bio-Rad's Argument

The 837 and 722 Patent Specification examples include an outlet placed at the bottom of the chamber to collect aqueous droplets in mineral oil:

The selected oil is not only immiscible with the droplets, but also has a density different from that of the droplets. The density differential between the selected oil and the droplets facilitates the movement of the droplets in an upward or downward direction where they can be collected. For example, aqueous droplets placed in a chamber of fluorinated oil will tend to rise due to their lower density. Accordingly, an outlet for collecting the displaced droplets can be positioned at the top of the chamber. In another example, aqueous droplets placed in a chamber of certain mineral oils will tend to sink due to their greater density. Accordingly, an outlet for collecting the displaced droplets can be positioned at the bottom of the chamber.

IPR2020-00087 Ex. 1001 (837 Patent) at 4:4-16  
IPR2020-00089 Ex. 1001 (722 Patent) at 4:4-16



# Incubation Chamber: “separation chamber”

## The Specification Contradicts Bio-Rad’s Argument

The 837 and 722 Patent Specification examples include continued flowing of the immiscible fluid:

jars, and PCR tubes. In addition, the outlet may be connected to a second vessel, such that the continued flowing of the immiscible fluid eventually displaces the droplets from the chamber into the vessel.

In certain embodiments, the outlet is connected to a channel. The continued flowing of the immiscible fluid into the vessel directs the droplets towards the outlet and subsequently into the connected channel where the droplets may be collected in the vessel.

IPR2020-00087 Ex. 1001 (837 Patent) at 53:38-46

IPR2020-00089 Ex. 1001 (722 Patent) at 53:47-55



# Incubation Chamber: 3 Disputes (independent claims)

- (1) “separation chamber” (837 Patent & 722 Patent)
- (2) “droplet receiving outlet” (837 Patent)
  - Bio-Rad argues the incubation chamber does not have a “droplet receiving outlet”
  - Bio-Rad’s construction of “droplet receiving outlet” is “portion of the separation chamber where droplets are collected”
- (3) “sample” (837 Patent & 722 Patent) (not argued in sur-reply)

IPR2020-00087 POR at 25-31, 37-41, Pet. Reply at 12-14, Petition at 46-48

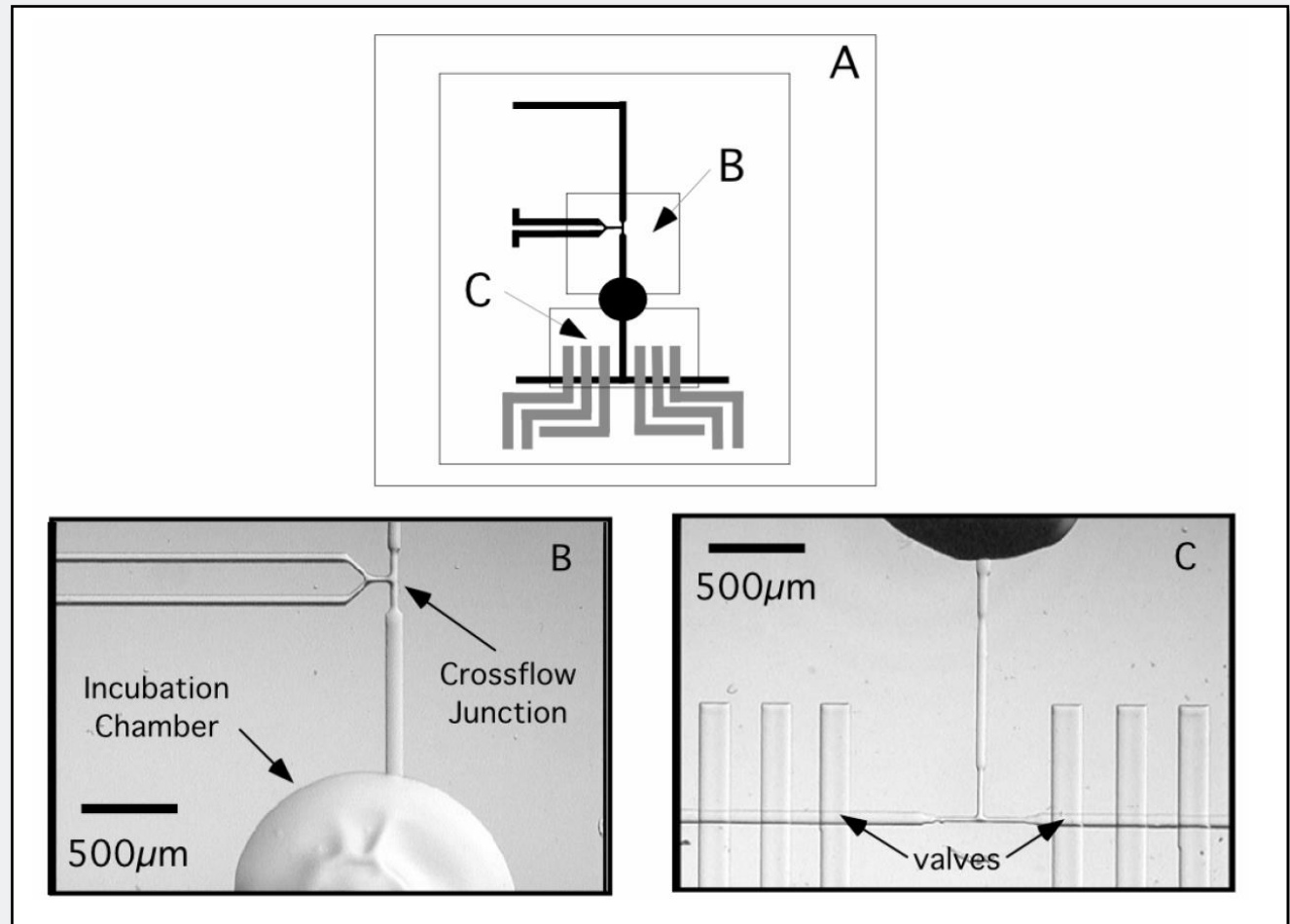
# Incubation Chamber: “droplet receiving outlet”

## **PO’s construction:**

“portion of the separation chamber where droplets are collected”

## **837 Patent [1.3]:**

“... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet.”



IPR2020-00087 Ex. 1003 (Thorsen) at 101 Fig. 3.28, Petition at 45-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 101 Fig. 3.28, Petition at 44-47

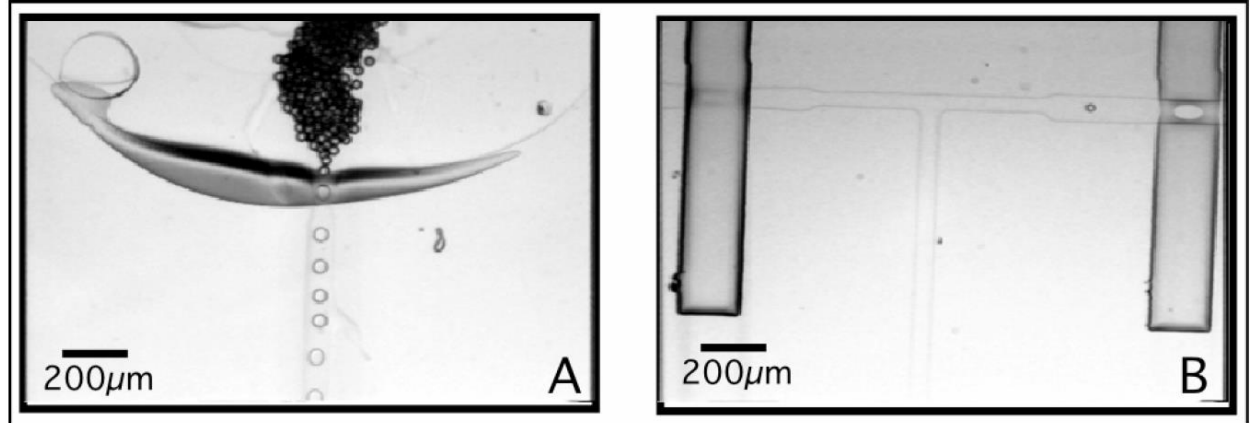
# Incubation Chamber: "droplet receiving outlet"

## PO's construction:

"portion of the separation chamber where droplets are collected"

## 837 Patent [1.3]:

"... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet."



**Figure 3.29:** A) Water droplets entering incubation cavity of Sylgard 184 device.  
B) Sorting of droplets using elastomeric valves.

Generated water droplets entered the "incubation" cavity, where they encountered a sharp drop in flow velocity (Figure 3.29A). After ~ 1 hour, the droplets had passed entirely through the cavity and re-entered the narrow (~60 μm wide) outlet channel, where they flowed toward the T-junction for valve-based sorting.

IPR2020-00087 Ex. 1003 (Thorsen) at 103 Fig. 3.29, Petition at 45-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 103 Fig. 3.29, Petition at 44-47

# Incubation Chamber: “droplet receiving outlet”

## **PO's construction:**

“portion of the separation chamber where droplets are collected”

## **837 Patent [1.3]:**

“... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet.”

reversed (<1 V green, +4 V red). Droplet frequency passing through the detection region was low, with a single droplet or small cluster of droplets leaving the microcavity and passing through the region every few seconds. Sorting efficiency was measured both by looking at the PMT output values of the output material in the collection wells. As droplets formed a large aggregate in the output wells, PMT readings were not especially useful as neighbor droplets washed out the signal from improperly sorted droplets. Visual inspection of the output wells was carried under a 40x objective (0.65NA) on an inverted microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. Of the thousands of sorted droplets in each well, only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00087 Ex. 1003 (Thorsen) at 111, Petition at 45-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 111, Petition at 44-47

# Incubation Chamber: “droplet receiving outlet”

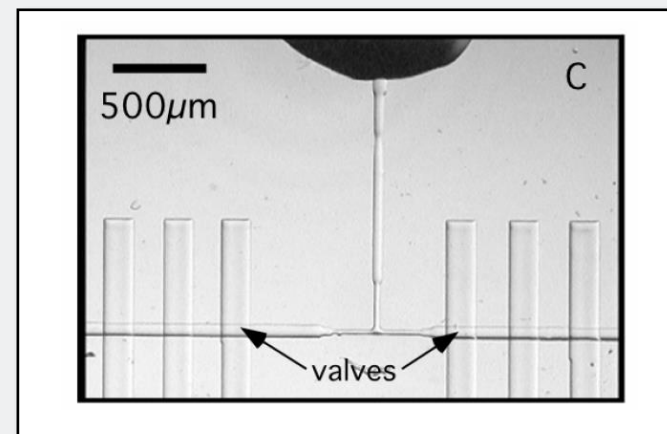


**Dr. Richard Fair**



“Under Bio-Rad’s interpretation of the claims, the ‘Incubation Chamber’ has both a droplet receiving chamber inlet where the microchannel delivers the droplets formed at the ‘Crossflow Junction’ and a droplet receiving outlet where the droplets exit the ‘Incubation Chamber’ for further processing at the ‘sorting junction.’ [Thorsen], 101.... Figure 3.28C is a ‘High resolution [image] of [the] sorting junction in chip showing valve structures’ and the droplet receiving outlet. *Id.* The droplets exit through this outlet to the sorting junction....

...  
Thorsen also discloses that the droplets will collect in the well. *Id.* 110-111.... Using a pipette to remove droplets from an open well would be routine and well-known to a person of ordinary skill in the art.”



IPR2020-00087 Ex. 1008 at ¶¶ 128-129; Ex. 1003 (Thorsen) at Fig. 3.28C



# Incubation Chamber: “droplet receiving outlet”

## 837 and 722 Patent Specification:

jars, and PCR tubes. In addition, the outlet may be connected to a second vessel, such that the continued flowing of the immiscible fluid eventually displaces the droplets from the chamber into the vessel.

In certain embodiments, the outlet is connected to a channel. The continued flowing of the immiscible fluid into the vessel directs the droplets towards the outlet and subsequently into the connected channel where the droplets may be collected in the vessel.

IPR2020-00087 Ex. 1001 (837 Patent) at 53:38-46  
IPR2020-00089 Ex. 1001 (722 Patent) at 53:47-55

# Incubation Chamber: 3 Disputes (independent claims)

- (1) “separation chamber” (837 Patent & 722 Patent)
- (2) “droplet receiving outlet” (837 Patent)
- (3) “sample” (837 Patent & 722 Patent)
  - Bio-Rad dropped its “sample”-related argument from its sur-reply as to Thorsen’s incubation chamber
  - In its POR, Bio-Rad argued red/green dyes were not “samples”
  - Bio-Rad’s construction of “sample” is “fluid containing molecules, cells, small molecules (organic or inorganic) or particles”

IPR2020-00087 POR at 25-31, 37-41, Pet. Reply at 5-7, Petition at 33-45, PO Sur-Reply at 4-7  
IPR2020-00089 POR at 23-27, Pet. Reply at 4-6, Petition at 28-41, PO Sur-Reply at 4-7

# Incubation Chamber: “sample”-related limitations

## PO’s construction:

“fluid containing molecules, cells, small molecules (organic or inorganic) or particles”

## 837 Patent [1.2]:

“... at least one droplet formation module comprising a **sample** inlet, an immiscible fluid inlet, and a junction ... configured to produce droplets comprising the **sample** surrounded by the immiscible fluid;”

## 722 Patent [1.1]/[14.1], [1.3]/[14.3]:

“providing an aqueous fluid comprising a **sample** through a **sample** inlet ... partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a **sample**;”

mineral oil and its elastomeric properties. A significant design modification was added to the device to accommodate chemical screening reactions where incubations of several minutes to a few hours are optimal (Figure 3.28A). The serpentine output channel at the crossflow junction was replaced with a large (~500  $\mu\text{m}$ ) high cavity to slow down the droplets and create an on-chip incubation chamber (Figure 3.28B). This design element not only created a more flexible on-chip screening environment, but also significantly reduced the footprint of the chip from 1 in<sup>2</sup> to 0.5 in<sup>2</sup>. A control layer of valves was added

IPR2020-00087 Ex. 1003 (Thorsen) at 99, Petition at 30-37  
IPR2020-00089 Ex. 1003 (Thorsen) at 99, Petition at 29-34, 43-44

# Incubation Chamber: “sample”-related limitations

## PO’s construction:

“fluid containing molecules, cells, small molecules (organic or inorganic) or particles”

## 837 Patent [1.2]:

“... at least one droplet formation module comprising a **sample** inlet, an immiscible fluid inlet, and a junction ... configured to produce droplets comprising the **sample** surrounded by the immiscible fluid;”

## 722 Patent [1.1]/[14.1], [1.3]/[14.3]:

“providing an aqueous fluid comprising a **sample** through a **sample** inlet ... partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a **sample**;”

### *Two Color Droplet Sorting*

As a functional demonstration of the device, the fluorescence-activated droplet sorter was used to sort 2-3 $\mu$ m diameter droplets filled with either green (fluorescein) or red (R-phycoerythrin) fluorescent dyes. Both dyes are excited by the 473nm blue laser and have appropriate distinct emission spectra for the two PMT detectors.

A Sylgard 184 microcavity crossflow device was primed with mineral oil/ 1% span 80 at the oil input, with fluorescein (130  $\mu$ M in PBS, pH 7.2) and R-phycoerythrin (830nM in 0.1M sodium phosphate buffer, 0.1 M NaCl, pH 7.4). Flow was balanced at the crossflow junction at 14.2/18.0 psi (w/o-s), with the oil physically separating the two dye channels (Figure 3.32A). Single color-droplets were generated by adjusting the

IPR2020-00087 Ex. 1003 (Thorsen) at 109-110, Petition at 30-37  
IPR2020-00089 Ex. 1003 (Thorsen) at 109-110, Petition at 29-34, 43-44

# Incubation Chamber: “sample”-related limitations

## PO’s construction:

“fluid containing molecules, cells, small molecules (organic or inorganic) or particles”

## 837 Patent [1.2]:

“... at least one droplet formation module comprising a **sample** inlet, an immiscible fluid inlet, and a junction ... configured to produce droplets comprising the **sample** surrounded by the immiscible fluid;”

## 722 Patent [1.1]/[14.1], [1.3]/[14.3]:

“providing an aqueous fluid comprising a **sample** through a **sample** inlet ... partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a **sample**;”

dye channels (Figure 3.32A). Single color-droplets were generated by adjusting the relative pressures of the two dye streams, allowing only one stream to enter the crossflow region for droplet formation while the other stream is blocked and isolated by a plug of oil (Figure 3.32B,C). As switching between the two dye streams was accomplished manually, several hundred droplets were generated for each color type each time the relative water pressures were changed. Using this process, thousands of fluorescein/R-phycoerythrin droplets were generated with a rough population distribution of ~1:1. The formed droplets entered the microcavity, where they slowed down and formed a mixed cluster of droplets.

IPR2020-00087 Ex. 1003 (Thorsen) at 109-110, Petition at 30-37  
IPR2020-00089 Ex. 1003 (Thorsen) at 109-110, Petition at 29-34, 43-44



# Incubation Chamber: “sample”-related limitations

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“providing an aqueous fluid comprising a **sample** through a **sample** inlet ... partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a **sample**;”

reversed (<1 V green, +4 V red). Droplet frequency passing through the detection region was low, with a single droplet or small cluster of droplets leaving the microcavity and passing through the region every few seconds. Sorting efficiency was measured both by looking at the PMT output values of the output material in the collection wells. As droplets formed a large aggregate in the output wells, PMT readings were not especially useful as neighbor droplets washed out the signal from improperly sorted droplets. Visual inspection of the output wells was carried under a 40x objective (0.65NA) on an inverted microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. Of the thousands of sorted droplets in each well, only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00087 Ex. 1003 (Thorsen) at 111, Petition at 30-37  
IPR2020-00089 Ex. 1003 (Thorsen) at 111, Petition at 29-34, 43-44

# Incubation Chamber: “sample”-related limitations

## Institution Decision

### Institution Decisions found that Thorsen discloses the “sample”-related limitations of the claims:

Second, Patent Owner’s argument fails to address the full scope of Petitioner’s argument regarding Thorsen Chapter 3. Petitioner asserts that a POSA would understand Thorsen’s incubation chamber system is for chemical screening reactions, including the screening of a sample, “such as a compound, composition and/or mixture of interest from any suitable source.” Pet. 33–35 (citing Ex. 1003, 10, 21, 26, 99; Ex. 1008 ¶¶ 106–109). Petitioner also asserts that Thorsen Chapter 3 discloses the use of the incubation chamber system to sort droplets containing red or green fluorescent dye. See Pet. 36–37 (citing Ex. 1003, 109–112).

For these reasons, and for purposes of institution, we are persuaded that Petitioner has sufficiently shown that Thorsen Chapter 3 discloses a “sample inlet” and “droplets comprising the sample” as recited in claim 1.

IPR2020-00087 Paper 9 (Institution Decision) at 34  
IPR2020-00089 Paper 8 (Institution Decision) at 29 (similar)

# Incubation Chamber: “separation chamber” Bio-Rad’s Argument

Bio-Rad’s argument is conclusory and unsupported:

droplets.” (089 Pet. at 34). Although fluorescein and R-phycoerythrin complexes may be used to indicate the presence of certain bacteria or antibodies, they are not “samples” themselves within the meaning of the ‘722 claims. (Ex. 2016 at ¶112).

112. I also do not agree with the assertion that Thorsen discloses the “sample” limitations because Thorsen “describes the use of the incubation chamber system to sort red vs. green droplets based on the presence of either ‘green (fluorescein) or red (R-phycoerythrin) fluorescent dyes’ contained in the droplets.” (Pet. at 36; Ex. 1008 ¶ 109). Although fluorescein and R-phycoerythrin complexes may be used to indicate the presence of certain bacteria or antibodies, they are not “samples” themselves within the meaning of the ‘722 claims.

IPR2020-00089 POR at 24-25, Ex. 2016 (Anna) at ¶ 112;  
*see also* IPR2020-00087 POR at 27, Ex. 2016 (Anna) at ¶ 122 (similar)

# Outlet Wells: 4 Disputes (independent claims)

Patent Owner raises only four disputes as to Thorsen's outlet wells for the independent claims:

- (1) "separation chamber" size and orientation-related limitations (837 Patent & 722 Patent)
- (2) Bio-Rad argues droplets and oil do not separate in Thorsen's outlet wells (837 Patent & 722 Patent)
- (3) "sample" (837 Patent & 722 Patent)
- (4) "droplet receiving outlet" (837 Patent)

IPR2020-00087 POR at 25-29, 32-37, 44-46, Pet. Reply at 14-29

IPR2020-00089 POR at 23-25, 28-35, Pet. Reply at 10-25

# Outlet Wells: 4 Disputes (independent claims)

- (1) “separation chamber” (837 Patent & 722 Patent)
  - Bio-Rad disputes whether the outlet wells meet the size and orientation requirements of the claimed “separation chamber”
  - Bio-Rad’s construction of “separation chamber” is “chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”
- (2) Bio-Rad argues droplets do not separate from oil in Thorsen’s outlet wells (837 Patent & 722 Patent)
- (3) “sample” (837 Patent & 722 Patent)
- (4) “droplet receiving outlet” (837 Patent)

IPR2020-00087 POR at 25-29, 32-37, 44-46, Pet. Reply at 15-22, Petition at 52-53, 56-58  
IPR2020-00089 POR at 23-25, 28-35, Pet. Reply at 11-19, Petition at 46-47, 51-53, 55-56



# Outlet Wells: “separation chamber” size & orientation

## **PO's construction:**

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.4], [1.6], [1.7]:**

“[1.4] wherein the separation chamber is upright to the microchannel and out of the horizontal plane; ...

[1.6] wherein the separation chamber has a wider cross-section than the microchannel cross-section to accumulate a plurality of droplets comprising the sample and [1.7] is of a volume sufficient to separate the plurality of droplets ... from the immiscible fluid within the separation chamber.”

## **722 Patent [1.5]/[14.5], [1.6]/[14.6]:**

“[1.5] wherein the separation chamber has a wider cross-section than the main channel cross-section and

[1.6] the separation chamber is disposed perpendicular to the main channel;”

reversed (<1 V green, +4 V red). Droplet frequency passing through the detection region

was low, with a single droplet or small cluster of droplets leaving the microcavity and

passing through the region every few seconds. Sorting efficiency was measured both by

looking at the PMT output values of the output material in the collection wells. As

droplets formed a large aggregate in the output wells, PMT readings were not especially

useful as neighbor droplets washed out the signal from improperly sorted droplets. Visual

inspection of the output wells was carried under a 40x objective (0.65NA) on an inverted

microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly

scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds

25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. Of the thousands

of sorted droplets in each well, only a small fraction (~1-2%) were of the wrong dye type.

Thorsen at 111; IPR2020-00087 Petition at 52-53, 56-58, Pet. Reply at 16-22;  
IPR2020-00089 Petition at 46-47, 51-53, 55-56, Pet. Reply at 12-19

# Outlet Wells: “separation chamber” size & orientation

## PO's construction:

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## 837 Patent [1.4], [1.6], [1.7]:

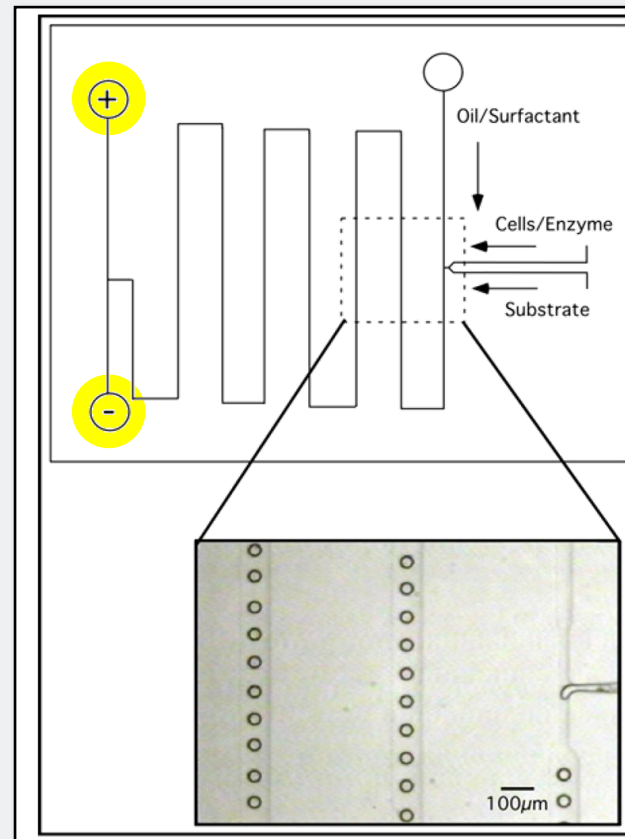
“[1.4] wherein the separation chamber is upright to the microchannel and out of the horizontal plane; ...

[1.6] wherein the separation chamber has a wider cross-section than the microchannel cross-section to accumulate a plurality of droplets comprising the sample and [1.7] is of a volume sufficient to separate the plurality of droplets ... from the immiscible fluid within the separation chamber.”

## 722 Patent [1.5]/[14.5], [1.6]/[14.6]:

“[1.5] wherein the separation chamber has a wider cross-section than the main channel cross-section and

[1.6] the separation chamber is disposed perpendicular to the main channel;”



**Figure 4.2:** Serpentine channel design for biochemical screening chip. Monodisperse water droplets in mineral oil/2% Span 80 retain a coherent pattern as they flow towards the device outlet.

Thorsen at 116, Fig. 4.2; IPR2020-00087 Petition at 52-53, 56-58, Pet. Reply at 16-22; IPR2020-00089 Petition at 46-47, 51-53, 55-56, Pet. Reply at 12-19

# Outlet Wells: “separation chamber” size & orientation

## PO's construction:

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## 837 Patent [1.4], [1.6], [1.7]:

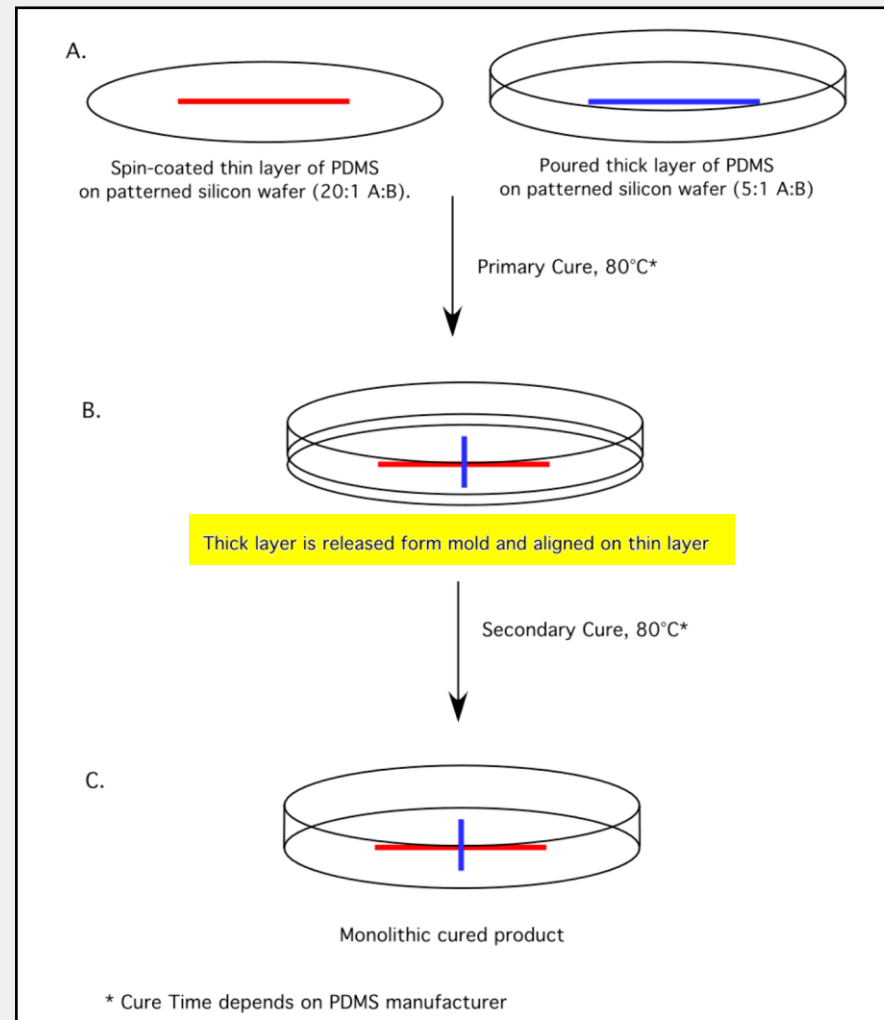
“[1.4] wherein the separation chamber is upright to the microchannel and out of the horizontal plane; ...

[1.6] wherein the separation chamber has a wider cross-section than the microchannel cross-section to accumulate a plurality of droplets comprising the sample and [1.7] is of a volume sufficient to separate the plurality of droplets ... from the immiscible fluid within the separation chamber.”

## 722 Patent [1.5]/[14.5], [1.6]/[14.6]:

“[1.5] wherein the separation chamber has a wider cross-section than the main channel cross-section and

[1.6] the separation chamber is disposed perpendicular to the main channel;”



Thorsen at 100, Fig. 3.27; IPR2020-00087 Petition at 29-30, 52-53, 56-58, Pet. Reply at 16-22; IPR2020-00089 Petition at 41-43, 46-47, 51-53, 55-56, Pet. Reply at 12-19

# Outlet Wells: “separation chamber” size & orientation

## **PO’s construction:**

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.4], [1.6], [1.7]:**

“[1.4] wherein the separation chamber is upright to the microchannel and out of the horizontal plane; ...

[1.6] wherein the separation chamber has a wider cross-section than the microchannel cross-section to accumulate a plurality of droplets comprising the sample and [1.7] is of a volume sufficient to separate the plurality of droplets ... from the immiscible fluid within the separation chamber.”

## **722 Patent [1.5]/[14.5], [1.6]/[14.6]:**

“[1.5] wherein the separation chamber has a wider cross-section than the main channel cross-section and

[1.6] the separation chamber is disposed perpendicular to the main channel;”

**Bio-Rad admits Thorsen’s outlets are formed using a punch to bore through the PDMS layers:**

“Thorsen fabricated the openings in his devices by “punching” holes in the microchip itself at the channel ends, as was standard practice at the time.”

IPR2020-00087 POR at 2; IPR2020-00089 POR at 2



# Outlet Wells: “separation chamber” size & orientation



**Dr. Richard Fair**



Dr. Fair explained that multiple disclosures from Thorsen confirm that the outlet wells meet the size and orientation limitations of the “separation chamber”:

- Thorsen discloses that the outlet wells are large enough to accommodate “thousands” of droplets. ¶¶ 140, 142.
- Thorsen’s fabrication technique confirms that outlet wells large enough to hold thousands of droplets will extend upright, out of the plane of, and perpendicular to the channels. ¶¶ 140, 142.
- A POSA would understand “outlet well” “to refer to an open-topped, bucket-like structure from which the accumulated droplets could be extracted, e.g., by using a micropipette” which was routine in the art. ¶¶ 141, 129-130, 83, 108.
- A POSA would understand Thorsen’s outlet well to have a volume of at least 0.1  $\mu\text{l}$  (the minimum volume compatible with a micropipette), sufficient to hold >30,000 droplets. ¶ 143.

IPR2020-00087 Ex. 1008 (Fair) at ¶¶ 83, 108, 129-130, 140-143, Petition at 52-58  
*see also* IPR2020-00089 Ex. 1008 at ¶¶ 85, 106, 147-151, Petition at 46-47, 51-56 (similar)



# Outlet Wells: “separation chamber” size & orientation Institution Decision

Institution Decisions found the outlet wells disclose the size & orientation-related limitations of the “separation chamber”:

After considering both parties’ arguments, we are persuaded that Petitioner’s arguments and evidence concerning claim element 1.4 meet the threshold for institution. We are persuaded in particular by Petitioner’s analysis and Dr. Fair’s testimony regarding Thorsen’s disclosure of how the channels and wells are formed. Pet. 52; Ex. 1003, 102; Ex. 1008 ¶¶ 140, 141. We are also persuaded by Petitioner’s analysis and Dr. Fair’s testimony pertaining to Thorsen’s disclosure that “thousands” of droplets are collected “in each well.” Pet. 52; Ex. 1003, 111; Ex. 1008 ¶¶ 140, 142. We

IPR2020-00087 Paper 9 (Institution Decision) at 41  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 37 (similar)

# Outlet Wells: “separation chamber” size & orientation Institution Decision

Institution Decisions found the outlet wells disclose the size & orientation-related limitations of the “separation chamber”:

collected “in each well.” Pet. 52; Ex. 1003, 111; Ex. 1008 ¶¶ 140, 142. We are also persuaded by Petitioner’s analysis and Dr. Fair’s testimony regarding a POSA’s understanding of Thorsen’s outlet well as a structure from which accumulated droplets can be extracted using a micropipette. Pet. 52–53; Ex. 1008, ¶¶ 141, 143. Patent Owner’s counterargument raises a factual dispute that is best resolved upon a full record. We note that at this stage of the proceeding, Patent Owner does not point to evidence that “the outlet well may have the same height as the main channel” (Prelim. Resp. 27), and we find Patent Owner’s argument is not supported by Thorsen’s description of how the devices are designed and fabricated. Ex. 1003, 98–102.

IPR2020-00087 Paper 9 (Institution Decision) at 41  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 37 (similar)

# Outlet Wells: “separation chamber” size & orientation Institution Decision

Institution Decisions found the outlet wells disclose the size & orientation-related limitations of the “separation chamber”:

After considering both parties’ arguments, we are persuaded that Petitioner’s arguments and evidence concerning claim element 1.6 meet the threshold for institution. We are persuaded in particular by the Petitioner’s analysis pertaining to the number of droplets that accumulate in Thorsen’s outlet wells. Pet. 56; Ex. 1003, 111. Patent Owner’s counterargument is contradicted by its subsequent argument that “the outlet well may have the same height as the main channel but a wider cross-section.” Prelim. Resp. 27.

IPR2020-00087 Paper 9 (Institution Decision) at 42  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 38 (similar)

# Outlet Wells: “separation chamber” size & orientation Institution Decision

Institution Decisions found the outlet wells disclose the size & orientation-related limitations of the “separation chamber”:

After considering both parties’ arguments, we are persuaded that Petitioner has presented sufficient arguments and evidence concerning outlet wells and claim element 1.7 to meet the threshold for institution. For example, we are persuaded in particular by Dr. Fair’s testimony that a POSA will understand Thorsen’s outlet well to have a volume greater than 0.1  $\mu\text{l}$ , which is sufficient to accommodate at least ~30,000 of Thorsen’s droplets and “plenty of room for thousands or even ten thousand droplets to separate from the oil.” Ex. 1008 ¶¶ 130, 155. Dr. Fair bases that volume on the minimum volume that can be collected by micropipette. *Id.* ¶¶ 141, 143. At

IPR2020-00087 Paper 9 (Institution Decision) at 42-43  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 38-39 (similar)



# Outlet Wells: “separation chamber” size & orientation

## Bio-Rad’s Argument

Bio-Rad’s post-institution arguments fail to address the flaws recognized by this Board at Institution:

- Bio-Rad repeats its argument that the outlet wells could be in the same plane as the microchannels as PO’s only explanation for how Thorsen’s outlet wells could accommodate thousands of droplets without meeting the “separation chamber” size & orientation limitations.
- Bio-Rad still identifies no evidence in Thorsen supporting its “same plane” theory.

IPR2020-00087 POR at 33-34, 45-46

IPR2020-00089 POR at 29-30, 34

- The Petition, Petitioner’s Reply, and Dr. Fair explain why an outlet well “in the same plane” as the microchannels would be contrary to Thorsen’s disclosures and incompatible with Thorsen’s system.

IPR2020-00087 Petition at 32-33, 52-53, Ex. 1008 (Fair) at ¶¶ 140-143, Pet. Reply at 22, Ex. 1072 (Fair2) at ¶¶ 34-39

IPR2020-00089 Petition at 30-31, 52-53, Ex. 1008 (Fair) at ¶¶ 147-151, Pet. Reply at 18-19, Ex. 1074 (Fair2) at ¶¶ 32-37



# Outlet Wells: “separation chamber” size & orientation Bio-Rad’s Argument Is Incompatible With Thorsen



**Dr. Richard Fair**



## Bio-Rad’s “in the same plane” theory:

“is directly contradicted by Thorsen’s fabrication teachings ... which describe that outlet wells have to be cut through the ‘thick’ layer in order to reach the ‘thin’ layer of microchannels. Thorsen, 98-102. In addition ... In Thorsen’s system, cutting a well with the same height as the microchannel would thus mean the microchannel height also extends through the entire PDMS layer(s), leaving the microchannel open at the top. Thorsen, 98-102. Because Thorsen uses pressure-driven flows of fluids in the microchannels to generate plugs/droplets, a person of ordinary skill in the art would know the microchannels cannot be open at the top, else applying pressure would spill fluids out the tops of the channels and make the system inoperable....”

IPR2020-00087 Ex. 1072 (Fair2) at ¶ 39, Petition at 32-33, 52-53, Pet. Reply at 22;  
IPR2020-00089 Ex. 1074 (Fair2) at ¶ 37, Petition at 30-31, 52-53, Pet. Reply at 18-19

# Outlet Wells: 4 Disputes (independent claims)

- (1) “separation chamber” size and orientation-related limitations (837 Patent & 722 Patent)
- (2) Bio-Rad argues droplets and oil do not separate in Thorsen’s outlet wells (837 Patent & 722 Patent)
  - Bio-Rad repeats its argument that 100% separation of droplets from oil is required
  - Bio-Rad’s argument contradicts its prior arguments to this Board and the District Court
- (3) “sample” (837 Patent & 722 Patent)
- (4) “droplet receiving outlet” (837 Patent)

IPR2020-00087 POR at 33, 37, Pet. Reply at 7-12, 15, Petition at 8-11, 57-58  
IPR2020-00089 POR at 29, 35, Pet. Reply at 6-10, 11-12, Petition at 8-11, 55-56

# Outlet Wells: “separation chamber”

## **PO’s construction:**

“a chamber sized to accommodate multiple droplets and an immiscible fluid in separated form”

## **837 Patent [1.7]:**

“...is of a volume sufficient to separate the plurality of droplets comprising the sample from the immiscible fluid within the separation chamber.”

## **722 Patent [1.7]/[14.7]:**

“separating the plurality of droplets from the immiscible fluid in the separation chamber based on the different densities of the droplets and the immiscible fluid.”

Thorsen’s outlet wells are large enough to accommodate “thousands” of droplets and immiscible fluid in separated form:

microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. Of the thousands of sorted droplets in each well, only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00087 Ex. 1003 (Thorsen) at 103, 111, Petition at 8-11, 57-58  
IPR2020-00089 Ex. 1003 (Thorsen) at 103, 111, Petition at 8-11, 55-56

# Outlet Wells: "separation chamber"



**Dr. Richard Fair**



"Thorsen discloses that the outlet well has a volume sufficient to accumulate 'thousands' of droplets (*id.*, 109-111, 115, Fig. 4.2) which again discloses a volume sufficient for the droplets to move relative to the oil phase such that the droplets will sink to the bottom of the outlet well while the mineral oil floats to the top, disclosing this limitation under Bio-Rad's interpretation.... As I discussed above in paragraphs 141-143, a person of ordinary skill in the art will understand Thorsen's disclosed outlet well to have a volume at least greater than 0.1  $\mu$ l. As I described in the previous paragraph, even a volume of only 0.1  $\mu$ l would be a sufficient volume to accommodate at least ~30,000 of Thorsen's droplets (plenty of room for thousands or even ten thousand droplets to separate from the oil under Bio-Rad's interpretation)."

IPR2020-00087 Ex. 1008 at ¶ 155  
*see also* IPR2020-00089 Ex. 1008 at ¶¶ 158, 161 (similar)

# Outlet Wells: “separation chamber”



**Dr. Richard Fair**



“It was also well-known to generate droplets using fluids of different densities... Aqueous (water) solutions and fluorinated oils have different densities... Water has a density of 1 g/cm<sup>3</sup>, while perfluorohexane and perfluorodecaline have densities of around 1.7 and 1.9 g/cm<sup>3</sup>, respectively. Ex. 1014, 4-5. These are constant, unchanging properties of these chemicals, and this difference in density means that the less dense water droplets necessarily will float to the top in denser oil. By contrast, mineral oil is less dense than water (between 0.8 and 0.9 g/ml), and so aqueous droplets would sink to the bottom. Ex. 1015, Ex. 1016, Ex. 1063, Ex. 1064.”

IPR2020-00087 Ex. 1008 at ¶ 51 (Section VI. State of the Art)

IPR2020-00089 Ex. 1008 at ¶ 51 (Section VI. State of the Art)



# Outlet Wells: “separation chamber” Institution Decision

Institution Decisions found that the outlet wells disclose the “separation chamber” and 722 Patent “separating” step:

After considering both parties’ arguments, we are persuaded that Petitioner has presented sufficient arguments and evidence concerning outlet wells and claim element 1.7 to meet the threshold for institution. For example, we are persuaded in particular by Dr. Fair’s testimony that a POSA will understand Thorsen’s outlet well to have a volume greater than 0.1  $\mu\text{l}$ , which is sufficient to accommodate at least ~30,000 of Thorsen’s droplets and “plenty of room for thousands or even ten thousand droplets to separate from the oil.” Ex. 1008 ¶¶ 130, 155. Dr. Fair bases that volume on the minimum volume that can be collected by micropipette. *Id.* ¶¶ 141, 143. At

IPR2020-00087 Paper 9 (Institution Decision) at 42-43  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 38-39 (similar)

# Outlet Wells: 4 Disputes (independent claims)

- (1) “separation chamber” size and orientation-related limitations (837 Patent & 722 Patent)
- (2) Bio-Rad argues droplets and oil do not separate in Thorsen’s outlet wells (837 Patent & 722 Patent)
- (3) “sample” (837 Patent & 722 Patent)
  - Thorsen’s red/green dye test system included outlet wells and Bio-Rad does not dispute enablement for those dyes
  - Bio-Rad admits the cells in Thorsen’s Chapter 4 meet the “sample” limitations, but Bio-Rad disputes enablement
- (4) “droplet receiving outlet” (837 Patent)

IPR2020-00087 POR at 25-29, 48-50, Pet. Reply at 5-7, 15, 26-29, Petition at 14-15, 33-37

IPR2020-00089 POR at 23-25, 41-42, Pet. Reply at 4-6, 11, 22-25, Petition at 14-15, 29-34

# Outlet Wells: “sample”-related limitations

## Thorsen’s Red/Green Dye Sorting

### PO’s construction:

“fluid containing molecules, cells, small molecules (organic or inorganic) or particles”

### 837 Patent [1.2]:

“... at least one droplet formation module comprising a **sample** inlet, an immiscible fluid inlet, and a junction ... configured to produce droplets comprising the **sample** surrounded by the immiscible fluid;”

### 722 Patent [1.1]/[14.1], [1.3]/[14.3]:

“providing an aqueous fluid comprising a **sample** through a **sample** inlet ... partitioning the aqueous fluid with the immiscible fluid to form a plurality of droplets in the main channel, wherein at least one droplet comprises a **sample**;”

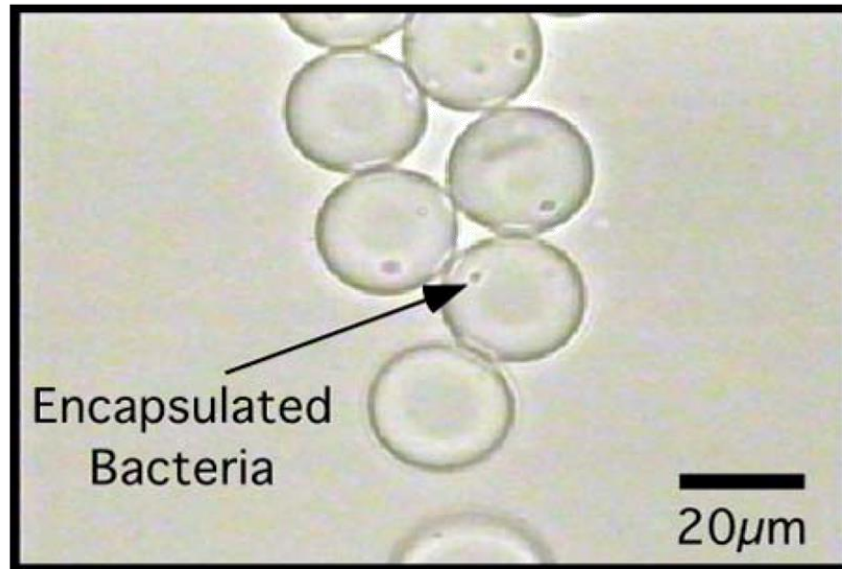
### *Two Color Droplet Sorting*

As a functional demonstration of the device, the fluorescence-activated droplet sorter was used to sort 2-3 $\mu$ m diameter droplets filled with either green (fluorescein) or red (R-phycoerythrin) fluorescent dyes. Both dyes are excited by the 473nm blue laser and have appropriate distinct emission spectra for the two PMT detectors.

microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. Of the thousands of sorted droplets in each well, only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00087 Ex. 1003 (Thorsen) at 109-111, Petition at 31-34  
IPR2020-00089 Ex. 1003 (Thorsen) at 109-111, Petition at 29-34, 43-44

# "sample"-related limitations: Thorsen's Chapter 4 Cell Assay System Is An Enabling Disclosure



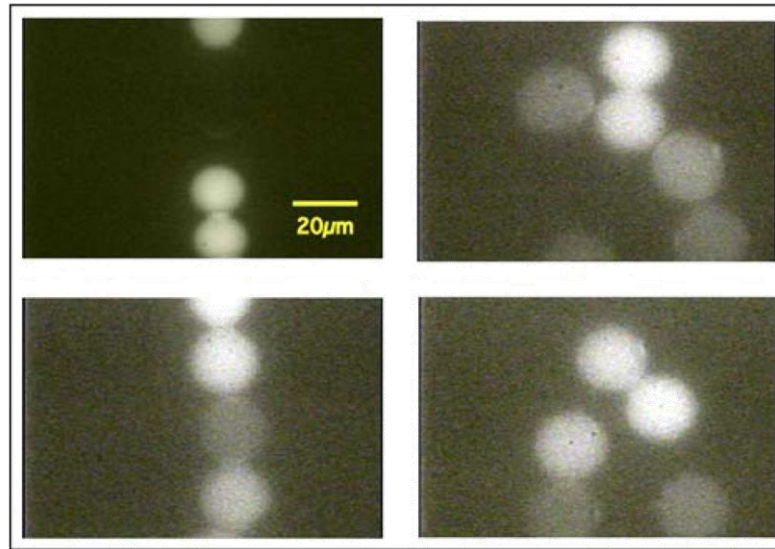
**Figure 4.4:** Encapsulated bacteria in droplets generated by microfluidic crossflow.

IPR2020-00087 Ex. 1003 (Thorsen) at 119, Fig. 4.4, Petition at 14-15  
IPR2020-00089 Ex. 1003 (Thorsen) at 119, Fig. 4.4, Petition at 14-15



# "sample"-related limitations: Thorsen's Chapter 4 Cell Assay System Is An Enabling Disclosure

“However, a few good examples of catalysis inside the formed droplets were still acquired (Figure 4.5).”



**Figure 4.5:** Monodisperse droplets containing *E. coli* expressing recombinant pNB esterase and fluorescein diacetate substrate. Droplets containing no cells are non-fluorescent while droplets containing one or more enzyme-expressing cells have converted the substrate into the fluorescent fluorescein product.

IPR2020-00087 Ex. 1003 (Thorsen) at 119-120, Fig. 4.5, Petition at 14-15  
IPR2020-00089 Ex. 1003 (Thorsen) at 119-120, Fig. 4.5, Petition at 14-15



# “sample”-related limitations: Thorsen’s Chapter 4 Cell Assay System Is An Enabling Disclosure

“Hundreds of crossflow devices were fabricated, injected with pNB esterase assay components, and ultimately discarded in an attempt to discover the perfect operating conditions. This section is intended to offer some insight into the troubleshooting process that evolved as the on-chip assay for pNB esterase was being developed.”

IPR2020-00087 Ex. 1003 (Thorsen) at 121, Petition at 14-15  
IPR2020-00089 Ex. 1003 (Thorsen) at 121, Petition at 14-15

# "sample"-related limitations: Thorsen's Chapter 4 Cell Assay System Is An Enabling Disclosure



**Dr. Richard Fair**



"Thorsen further created actual prototypes of these disclosed systems and obtained successful experimental results and photos confirming that his disclosed systems could in fact generate droplets (including droplets containing a sample of interest such as the *E. coli* cells in the recombinant enzyme assay), incubate those droplets in an incubation chamber and/or slow their travel through a serpentine outlet channel, sort the droplets using a system of valves, and distinguish between (fluorescent) droplets containing cells expressing the recombinant enzyme activity and (non-fluorescent) droplets that contained cells not expressing the recombinant enzyme activity. *E.g.*, [Thorsen], Figs. 3.28, 3.29, 4.1, 4.2, 4.4, 4.5, 111-112, 127, 108; *see also generally, id.*, 52-127."

IPR2020-00087 Ex. 1008 (Fair) at ¶ 63  
*see also* IPR2020-00089 Ex. 1008 (Fair) at ¶ 65 (similar)

# "sample"-related limitations: Thorsen's Chapter 4 Cell Assay System Is An Enabling Disclosure



**Dr. Richard Fair**



"Like the best scientists, Thorsen was also open about difficulties that he encountered in producing prototypes of his disclosed systems and obtaining example experimental results. He openly disclosed that he fabricated hundreds of prototype crossflow devices that were 'ultimately discarded in an attempt to discover the perfect operating conditions.' ... He also provides a detailed description of difficulties he encountered in connection with improving the fluorescent-based cell assay system and the detailed trouble shooting steps he took in order to still obtain some successful experimental results from those prototypes. *Id.*, 120-127. He disclosed expressly and unequivocally that he did in fact obtain successful experimental results from a prototype of that assay (*id.*, Figs. 4.4-4.5, 118-120) as well as from a fluorescence-based droplet sorting test using his incubation chamber system (*id.*, 109-111). He also encouraged future improvements by others in the field, including 'in areas such as surface chemistry technology and reagent stability.' *Id.*, 127."

IPR2020-00087 Ex. 1008 (Fair) at ¶ 64  
see also IPR2020-00089 Ex. 1008 (Fair) at ¶ 66 (similar)

# "sample"-related limitations: Chapter 4 Is Enabling Institution Decision

Institution Decisions found the independent claims do not require the use of any particular type of sample (e.g. cells):

At this stage of the proceeding, and in view of the presumption of prior art enablement, we are persuaded to grant institution notwithstanding Patent Owner's arguments concerning Thorsen's purported lack of success in using the disclosed device to conduct biochemical experiments. Patent Owner's arguments raise factual disputes that are best resolved upon a full record. Moreover, challenged claim 1 does not require that the assembly be capable of use with any particular type of sample, such as cells, enzymes, or other biological material. Accordingly, for these reasons and based on the record at this stage of the proceeding, we are persuaded that Petitioner's arguments and evidence regarding Thorsen Chapter 4 are sufficient to show disclosure of a "sample" as recited claim element 1.2, notwithstanding Patent Owner's arguments to the contrary.

IPR2020-00087 Paper 9 (Institution Decision) at 37-39  
*see also* IPR2020-00089 Paper 8 (Institution Decision) at 33-35 (similar)

# Outlet Wells: 4 Disputes (independent claims)

- (1) “separation chamber” size and orientation limitations
- (2) Bio-Rad argues droplets and oil do not separate in Thorsen’s outlet wells (837 Patent & 722 Patent)
- (3) “sample” (837 Patent & 722 Patent)
- (4) “droplet receiving outlet” (837 Patent)
  - Bio-Rad argues Thorsen does not disclose collecting droplets from an outlet well by micropipette
  - Bio-Rad’s claim construction requires only a “portion of the separation chamber where droplets are collected”
  - Bio-Rad does not dispute that collection of droplets from wells by micropipette was routine and well-known

IPR2020-00087 POR at 41-44, Pet. Reply at 22-25, Petition at 47-51



# Outlet Wells: “droplet receiving outlet” Droplets Are Collected In The Wells

## **PO's construction:**

“portion of the separation chamber where droplets are collected”

## **837 Patent [1.3]:**

“... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet.”

reversed (<1 V green, +4 V red). Droplet frequency passing through the detection region was low, with a single droplet or small cluster of droplets leaving the microcavity and passing through the region every few seconds. Sorting efficiency was measured both by looking at the PMT output values of the output material **in the collection wells.** As droplets formed a large aggregate in the output wells, PMT readings were not especially useful as neighbor droplets washed out the signal from improperly sorted droplets. Visual inspection of the output wells was carried under a 40x objective (0.65NA) on an inverted microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. **Of the thousands of sorted droplets in each well,** only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00087 Ex. 1003 (Thorsen) at 111, Petition at 47-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 111, Petition at 46-47, 57-58

# Outlet Wells: “droplet receiving outlet” Droplets Are Collected In The Wells

## **PO's construction:**

“portion of the separation chamber where droplets are collected”

## **837 Patent [1.3]:**

“... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet.”

from the photoresist molds of the patterns. With a 90  $\mu\text{m}$  wide x 10  $\mu\text{m}$  high output channel ~8 cm in length, formed droplets required ~20 minutes to reach the output well. Given adequate spacing between the monodisperse droplets at the crossflow junction, water droplets in mineral oil/2% Span 80 (13.0/15.0 psi w/o-s) traveled as a perfectly coherent stream from the crossflow junction to the outlet well (Figure 4.2). At higher

IPR2020-00087 Ex. 1003 (Thorsen) at 115, Petition at 47-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 115, Petition at 46-47, 57-58

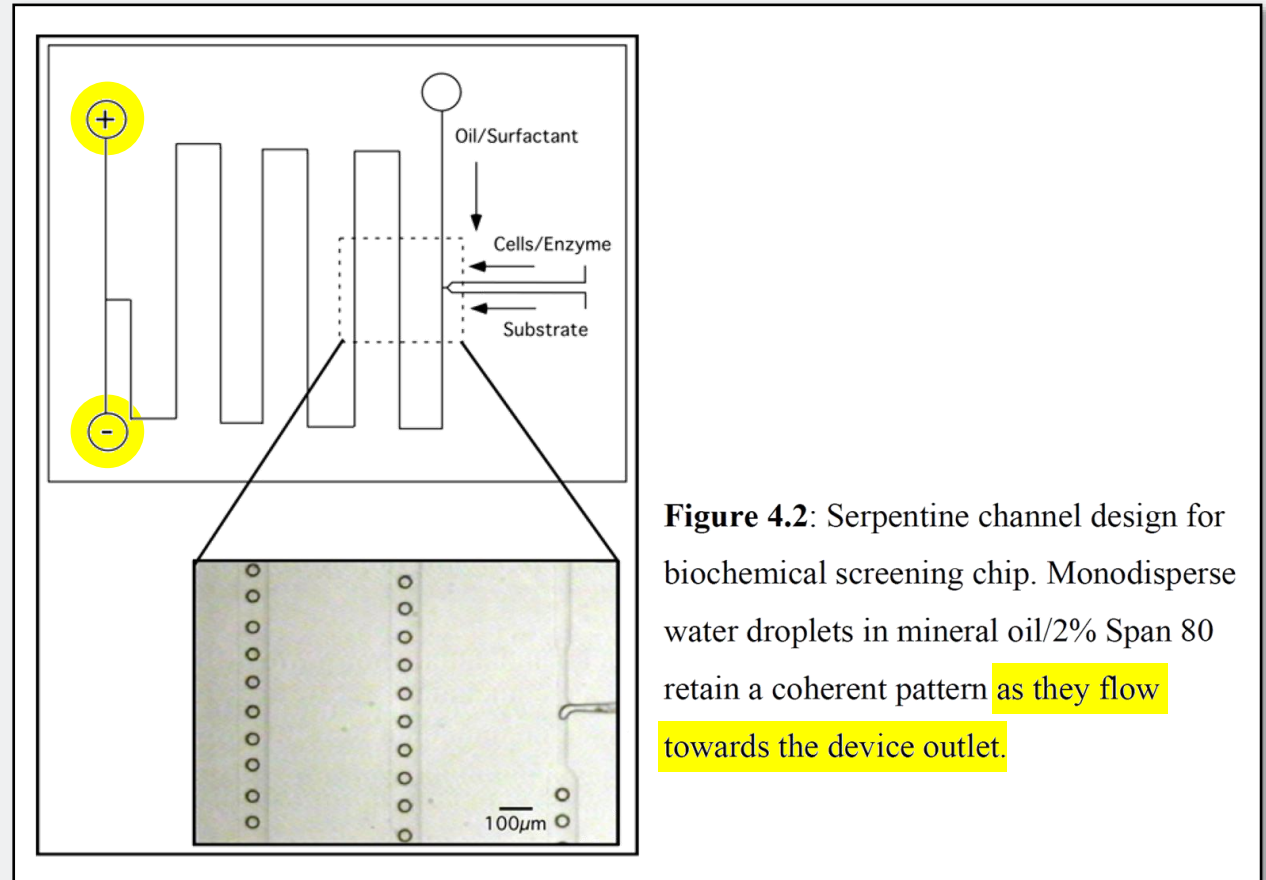
# Outlet Wells: “droplet receiving outlet” Droplets Are Collected In The Wells

## PO's construction:

“portion of the separation chamber where droplets are collected”

## 837 Patent [1.3]:

“... at least one downstream separation chamber comprising a droplet receiving chamber inlet and at least one droplet receiving outlet.”



**Figure 4.2:** Serpentine channel design for biochemical screening chip. Monodisperse water droplets in mineral oil/2% Span 80 retain a coherent pattern as they flow towards the device outlet.

IPR2020-00087 Ex. 1003 (Thorsen) at 115-116, Fig. 4.2, Petition at 47-48  
IPR2020-00089 Ex. 1003 (Thorsen) at 115-116, Fig. 4.2, Petition at 46-47, 57-58

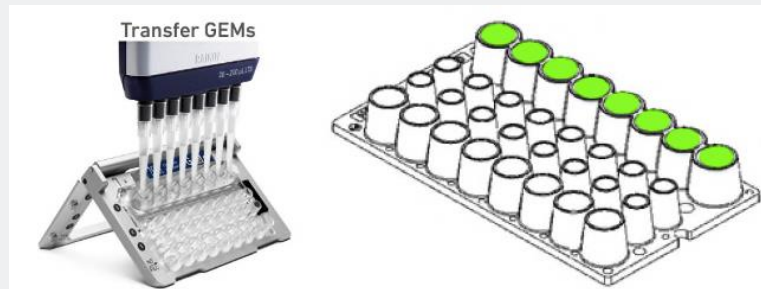
# Outlet Wells: “droplet receiving outlet” Droplets Are Collected In The Wells



**Dr. Richard Fair**



“Thorsen expressly discloses that the droplets will flow to the outlet well.... [Thorsen], 114-116, Figs. 4.1 and 4.2. Thorsen also discloses that the droplets will collect in the well. *Id.* 110-111... A [POSA] would know that because Thorsen’s water droplets are more dense than the mineral oil, Thorsen’s droplets will sink to the bottom of the outlet well. I understand that under Bio-Rad’s interpretation of the claims, an open well with droplets that have a different density than the immiscible fluid meets this claim limitation:



Ex. 1026, 11-12 (Bio-Rad’s infringement contentions identifying an open well as the separation chamber). Using a pipette to remove droplets from an open well would be routine and well-known to a person of ordinary skill in the art.”

IPR2020-00087 Ex. 1008 (Fair) at ¶ 129, Petition at 47-51, Pet. Reply at 22-23  
*see also* IPR2020-00089 Ex. 1008 (Fair) at ¶ 134 (similar), Petition at 46-47, 57-58



# Outlet Wells: “droplet receiving outlet” Bio-Rad’s Argument

- Bio-Rad argues that Thorsen does not disclose collecting droplets from the outlet wells using a micropipette.

IPR2020-00087 POR at 42  
IPR2020-00089 POR at 37 (similar)

- Bio-Rad’s argument ignores its own claim construction of “droplet receiving outlet”, which requires only a “portion of the separation chamber where droplets are collected.”
- Neither Bio-Rad nor its expert dispute that collection of droplets from outlet wells was routine and well-known.

IPR2020-00087 Ex. 1008 (Fair) at ¶¶ 83, 108, 129-136, Petition at 47-54, Pet. Reply at 22-23  
*see also* IPR2020-00089 Ex. 1008 (Fair) at ¶¶ 85, 106, 134-140, Petition at 57-61, Pet. Reply at 19-20 (similar)

# Dependent Claims

- Bio-Rad argues no additional disputes for the following dependent challenged claims, so these dependent claims rise or fall with the independent claims:
  - 722 Patent dependent claims 5-7, 10-13, 15-17
  - 837 Patent dependent claims 2-4, 10-13, 19
  
- Bio-Rad does argue additional disputes for these challenged dependent claims:
  - 722 Patent, claims 2-4
  - 837 Patent, claims 7-9

IPR2020-00087 POR at 46, 54-57, Pet. Reply at 5  
IPR2020-00089 POR at 35-39, 47-49, Pet. Reply at 4

# 722 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 722 Patent claims 2-4:

2. The method of claim 1, further comprising collecting the separated droplets.

3. The method of claim 1, wherein the separated droplets are collected in a vessel.

4. The method of claim 3, wherein the vessel is a PCR tube or a test tube.

# 722 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 722 Patent claims 2-4:

2. The method of claim 1, further comprising collecting the separated droplets.

3. The method of claim 1, wherein the separated droplets are collected in a vessel.

4. The method of claim 3, wherein the vessel is a PCR tube or a test tube.



# 722 Claim 2: “collecting the separated droplets” Thorsen Anticipates Claim 2

2. The method of claim 1, further comprising  
collecting the separated droplets.

- Bio-Rad repeats its same “no collecting” arguments addressed above for the 837 Patent’s “droplet receiving outlet” limitation.
- As addressed for the “droplet receiving outlet” dispute, Thorsen discloses collecting the separated droplets in the outlet wells (which are present in both Thorsen’s incubation chamber and serpentine channel systems).

IPR2020-00089 POR at 35-38

IPR2020-00089 Petition at 57-58, Pet. Reply at 19

# 722 Claim 2: “collecting the separated droplets” Thorsen Discloses Collecting The Separated Droplets

## 722 Patent Claim 2:

The method of claim 1, further comprising collecting the separated droplets.

reversed (<1 V green, +4 V red). Droplet frequency passing through the detection region was low, with a single droplet or small cluster of droplets leaving the microcavity and passing through the region every few seconds. Sorting efficiency was measured both by looking at the PMT output values of the output material **in the collection wells.** As droplets formed a large aggregate in the output wells, PMT readings were not especially useful as neighbor droplets washed out the signal from improperly sorted droplets. Visual inspection of the output wells was carried under a 40x objective (0.65NA) on an inverted microscope (Olympus IX50) equipped with a mercury lamp. The wells were rapidly scanned using both green (Ex 480 nm ds 30/ Em 535 nm ds 40) and red (Ex 540 nm ds 25/ Em 605 nm ds 55) filter sets to look for improperly sorted droplets. **Of the thousands of sorted droplets in each well,** only a small fraction (~1-2%) were of the wrong dye type.

IPR2020-00089 Ex. 1003 (Thorsen) at 111, Petition at 46-47, 57-58

# 722 Claim 2: “collecting the separated droplets” Thorsen Discloses Collecting The Separated Droplets



**Dr. Richard Fair**



“Thorsen expressly discloses that the droplets will flow ... to the outlet well.... [Thorsen 114-116], Figs. 4.1 and 4.2. Thorsen also discloses that the droplets will collect in the well. [Thorsen 110-111] ... A [POSA] would know from Thorsen’s disclosure that because Thorsen’s water droplets are more dense than the mineral oil, Thorsen’s droplets will sink to the bottom of the outlet well...”

IPR2020-00089 Ex. 1008 (Fair) at ¶¶ 134, 165, Petition at 46-47, 57-58

# 722 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 722 Patent claims 2-4:

2. The method of claim 1, further comprising collecting the separated droplets.

3. The method of claim 1, wherein the separated droplets are collected in a vessel.

4. The method of claim 3, wherein the vessel is a PCR tube or a test tube.



## 722 Claim 2: “collecting the separated droplets” Thorsen Anticipates Claim 3

3. The method of claim 1, wherein the separated droplets are collected in a vessel.

- Bio-Rad asserts without explanation or support that “neither Thorsen Chapter 3 nor 4 discloses collection of separated droplets in a ‘vessel’...”  
IPR2020-00089 POR at 38-39
- As addressed for the “droplet receiving outlet” dispute, Thorsen discloses collecting the separated droplets in the outlet wells which are vessels (and which are present in both Thorsen’s incubation chamber and serpentine channel systems).  
IPR2020-00089 Petition at 57-58, Pet. Reply at 19, Ex. 1008 (Fair) at ¶¶ 165-166

# 722 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 722 Patent claims 2-4:

2. The method of claim 1, further comprising collecting the separated droplets.

3. The method of claim 1, wherein the separated droplets are collected in a vessel.

4. The method of claim 3, wherein the vessel is a PCR tube or a test tube.

# 722 Claims 2-4: Thorsen Renders These Claims Obvious Alone Or w/ Ismagilov

- The Petition and Dr. Fair explain two independent bases for why Thorsen (alone or with Ismagilov) renders obvious 722 Claims 2-4:
  - (1) it would be obvious to a POSA to use a micropipette to move the droplets from Thorsen's outlet well to a PCR tube or a test tube
  - (2) it would be obvious to a POSA to substitute a PCR tube or a test tube in place of Thorsen's outlet well
- Both obviousness bases are motivated by Thorsen's disclosures about using his system for high-throughput chemical screening—once a POSA uses Thorsen's system to screen and sort desired from undesired droplets, a POSA will be motivated to collect those droplets in a PCR tube or test tube for further use.

IPR2020-00089 Petition at 21-22, 57-61, Pet. Reply at 19-22, 25-26, Ex. 1008 (Fair) at ¶¶ 165-174, Ex. 1074 (Fair2) at ¶¶ 25-31

# Thorsen Obviousness Grounds: High-Throughput Screening

High-throughput screening (HTS) is an established technology in the pharmaceutical industry<sup>26</sup>. Over the past decade, there has been a logarithmic increase in the industry's ability to screen large combinatorial libraries of compounds against target molecules<sup>27</sup>. The technology to achieve this has come in the form of robotics, high-

\* \* \*

The goal to develop assay systems that are smaller, faster, and cheaper has been realized in the field of microfluidics. Using materials such as glass, silicon, and hard plastics such as polycarbonate, microfluidic devices have been used to demonstrate a diverse array of biological applications, including biomolecular separations<sup>28</sup>, enzymatic assays<sup>29</sup>, polymerase chain reaction (PCR)<sup>30</sup>, and immunohybridization reactions<sup>31</sup>.

IPR2020-00087 Ex. 1003 (Thorsen) at 52, Petition at 21, 34, 62-66, Ex. 1008 (Fair) at ¶¶ 106-109, 167-176

IPR2020-00089 Petition at 21-22, 31, 57-61, Ex. 1008 (Fair) at ¶¶ 103-105, 165-174

# Thorsen Obviousness Grounds: High-Throughput Screening

The goal of this thesis is not to offer the solutions for specific screening problems, but rather to offer a general platform for addressing them. I describe the development of microfluidic devices for HTS applications having highly flexible parameters; a microencapsulation device suitable for aqueous solutions as well as organic solvents and a series of microfluidic devices with integrated elastomeric valves that function as high-density addressable arrays with picoliter volumes. Both of these systems create individual microenvironments with high informational content where thousands of single cells or small molecules such as DNA or proteins can be individually compartmentalized and examined. Model biochemical systems are presented to illustrate their potential as screening and selection tools in combinatorial assays such as recombinant enzyme libraries generated by random mutagenesis.

IPR2020-00087 Ex. 1003 (Thorsen) at 21, Petition at 21, 34, 62-66, Ex. 1008 (Fair) at ¶¶ 106-109, 167-176

IPR2020-00089 Petition at 21-22, 31, 57-61, Ex. 1008 (Fair) at ¶¶ 103-105, 165-174



# Thorsen Obviousness Grounds: Micropipettes



**Dr. Richard Fair**



"Thorsen describes using his system to encapsulate each bacteria cell in a droplet combined with a fluorescent marker that will fluoresce if the enzyme is present and then sorting the fluorescing droplets from the non-fluorescing droplets. *Id.* 116-120. The purpose of such a screening assay is to then collect the positive (fluorescent) droplets which will contain the bacteria cells with the desired mutation so that those bacteria cells of interest can then be used in future experiments. It thus would be obvious to a person of ordinary skill in the art reading Thorsen that the droplets could and would be collected from the outlet well, *e.g.* using a micropipette (a well-known technique as I discuss further in paragraph [159] below)."

IPR2020-00089 Ex. 1008 (Fair) at ¶ 106, Petition at 21-22, 57-61,  
Pet. Reply at 19-22, 25-26, Ex. 1074 (Fair2) at ¶¶ 25-31  
*see also* IPR2020-00087 Ex. 1008 (Fair) at ¶ 108 (similar), Petition at 21-22, 34-35, 62-63

# Thorsen Obviousness Grounds: Micropipettes



**Dr. Richard Fair**



"A micropipette (or microliter pipette) was a well-known, common piece of handheld laboratory equipment before the earliest possible filing date of the asserted patents. A micropipette is used to transfer a solution from one container to another container...."

IPR2020-00089 Ex. 1008 (Fair) at ¶ 159, Petition at 21-22, 57-61,  
Pet. Reply at 19-22, 25-26, Ex. 1074 (Fair2) at ¶¶ 25-31  
*see also* IPR2020-00087 Ex. 1008 (Fair) at ¶ 130 (similar), Petition at 21-22, 34-35, 62-63

# 837 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 837 Patent claims 7-9:

7. The assembly of claim 1, wherein the droplet receiving outlet is coupled to a vessel.

9. The assembly of claim 7, wherein the vessel is a PCR tube or a test tube.

8. The assembly of claim 7, wherein the droplet receiving outlet is sealably coupled to a vessel.

# 837 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 837 Patent claims 7-9:

7. The assembly of claim 1, wherein the droplet receiving outlet is coupled to a vessel.

9. The assembly of claim 7, wherein the vessel is a PCR tube or a test tube.

8. The assembly of claim 7, wherein the droplet receiving outlet is sealably coupled to a vessel.

# 837 Claims 7, 9: Thorsen Renders These Claims Obvious Alone Or w/ Ismagilov

- Bio-Rad does not dispute that its construction of “coupled” (“operably linked”) would be met by transferring droplets from the droplet receiving outlet to a vessel using either:
  - (i) a micropipette or
  - (ii) a microchannel connecting them

IPR2020-00087 POR at 23-24, Ex. 1075 (Anna Dep. Tr.) at 160:21-162:6, Pet. Reply at 3

- Thus, the disputes for 837 claims 7 and 9 are the same as those already addressed for 722 claims 2-4—namely, whether:
  - (1) it would be obvious to use a micropipette to move the droplets from Thorsen’s outlet well to a PCR tube or a test tube
  - (2) it would be obvious to substitute a PCR tube or a test tube in place of Thorsen’s outlet well at the end of the microchannel

IPR2020-00087 Petition at 21, 62-66, Pet. Reply at 3, 23-25, 29-30, Ex. 1008 (Fair) at ¶¶ 167-176, Ex. 1072 (Fair2) at ¶¶ 22, 27-33



# 837 Patent Dependent Claims – Additional Disputes

- Bio-Rad argues additional disputes for 837 Patent claims 7-9:

7. The assembly of claim 1, wherein the droplet receiving outlet is coupled to a vessel.

9. The assembly of claim 7, wherein the vessel is a PCR tube or a test tube.

8. The assembly of claim 7, wherein the droplet receiving outlet is **sealably coupled** to a vessel.

## 837 Claim 8: Ismagilov Confirms Sealably Coupling Outlets To Vessels Was Well-Known

- Bio-Rad does not raise any additional disputes for 837 Patent claim 8 beyond its arguments for claim 7.

IPR2020-00087 POR at 55-57

- Ismagilov confirms that it was well-known in the art that droplet receiving outlets (like Thorsen's) could be adapted for receiving a PCR tube or test tube in a sealable way.

IPR2020-00087 Petition at 21, 62-66, Pet. Reply at 3, 23-25, 29-30, Ex. 1008 (Fair) at ¶¶ 167-176, Ex. 1072 (Fair2) at ¶¶ 22, 27-33

## 837 Claim 8: Ismagilov Confirms Sealably Coupling Outlets To Vessels Was Well-Known

Ismagilov confirms a POSA would know (and Ismagilov discloses) that Thorsen's outlet can be adapted for receiving a test tube:

A group of manifolds (a region consisting of several channels that lead to or from a common channel) can be included to facilitate the movement of plugs from different analysis units, through the plurality of branch channels and to the appropriate solution outlet. Manifolds are preferably fabricated into the substrate at different depth levels. Thus, devices according to the invention may have a plurality of analysis units that can collect the solution from associated branch channels of each unit into a manifold, which routes the flow of solution to an outlet. The outlet can be adapted for receiving, for example, a segment of tubing or a sample tube, such as a standard 1.5 ml centrifuge tube. Collection can also be done using micropipettes.

IPR2020-00087 Ex. 1004 (Ismagilov 091) at 17:18-30, Petition at 21, 62-66, Pet. Reply at 3, 23-25, 29-30, Ex. 1008 (Fair) at ¶¶ 167-176, Ex. 1072 (Fair2) at ¶¶ 22, 27-33

## 837 Claim 8: Ismagilov Confirms Sealably Coupling Outlets To Vessels Was Well-Known

Ismagilov confirms that “leak-proof” techniques for sealably coupling containers to transfer droplets/plugs were well known:

[0191] In this experiment, the array of the plugs stored inside the Teflon tubing or the glass capillary (holding component) was transported into a PDMS microchannel (receiving component).

[0192] Three different configurations were used. In the first configuration, the inlet of the PDMS microchannel was coupled to a glass capillary obtained from Hampton research. This capillary was used as an adapter and it had a shape of a funnel. The small end of the capillary was inserted into the PDMS microchannel and the gap between the capillary and the wall of the PDMS microchannel was filled with half-cured PDMS. After incubation at 110° C. for 5 min, the half-cured PDMS was solidified and the junction became leak-proof. The capillary that had the array of the

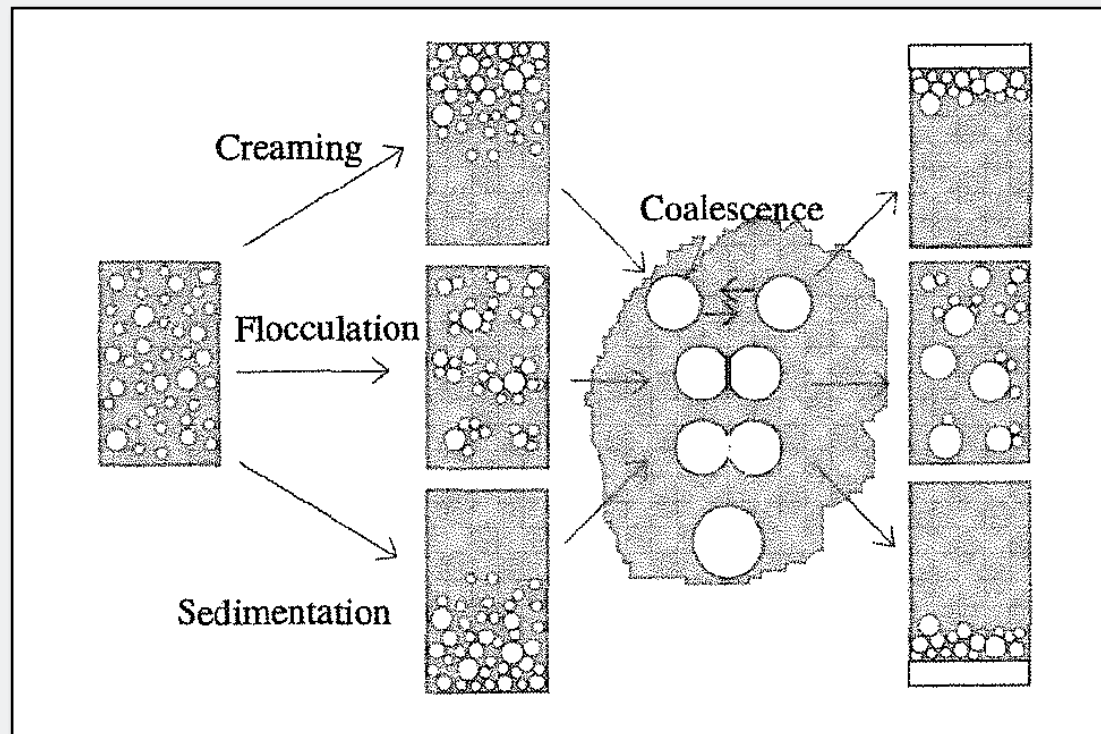
IPR2020-00087 Ex. 1005 (Ismagilov 119) at ¶¶ [0191]—[0192], Petition at 21, 62-66, Pet. Reply at 3, 23-25, 29-30, Ex. 1008 (Fair) at ¶¶ 167-176, Ex. 1072 (Fair2) at ¶¶ 22, 27-33



# Background State Of The Art: Density Separation

## Encyclopedic Handbook of Emulsion Technology (2001):

- Creaming and sedimentation of droplets based on density was well known



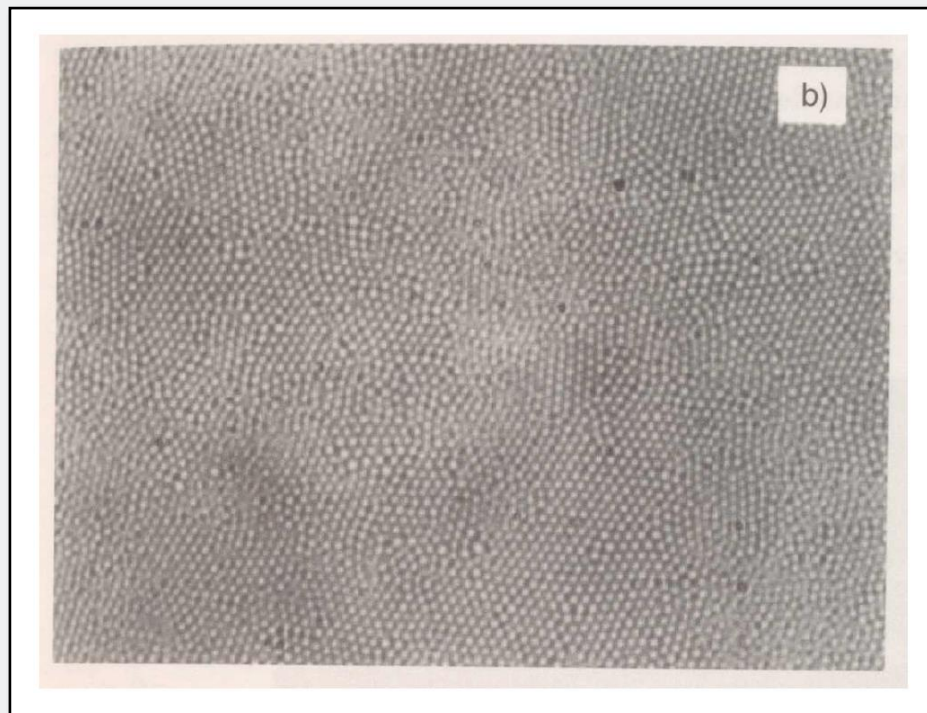
IPR2020-00087 Ex. 1009 at 51, Fig. 36, Petition at 8-9, Ex. 1008 (Fair) at ¶¶ 48-49  
IPR2020-00089 Ex. 1009 at 51, Fig. 36, Petition at 8-9, Ex. 1008 (Fair) at ¶¶ 48-49



# Background State Of The Art: Density Separation

## Emulsion Science: Basic Principles An Overview (2002):

- Collection of creamed/sedimented droplets was a well-known technique for producing monodisperse emulsions



IPR2020-00087 Ex. 1012 at 11-12, Fig. 3.1(b), Petition at 9-10, Ex. 1008 (Fair) at ¶ 50  
IPR2020-00089 Ex. 1012 at 11-12, Fig. 3.1(b), Petition at 9-10, Ex. 1008 (Fair) at ¶ 50