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Li et al.

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(54) **OPEN MICROFLUIDIC SYSTEM AND VARIOUS FUNCTIONAL ARRANGEMENTS THEREFORE**

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B01L 3/00 (2006.01)

(52) **U.S. Cl.**
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(58) **Field of Classification Search**
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See application file for complete search history.

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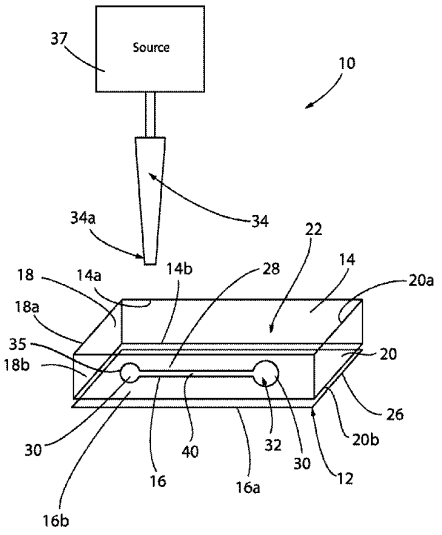
Primary Examiner — Jennifer Wecker
(74) *Attorney, Agent, or Firm* — Boyle Fredrickson, S.C.

(57) **ABSTRACT**

An open microfluidic system is provided. The open microfluidic system including the extreme wettability of exclusive liquid repellency (ELR), open microchannels with high lateral resolution and low profile, various valve arrangements, capable of a broad range flow rates, and capable of spatially and temporally trapping particles in open fluid.

23 Claims, 7 Drawing Sheets



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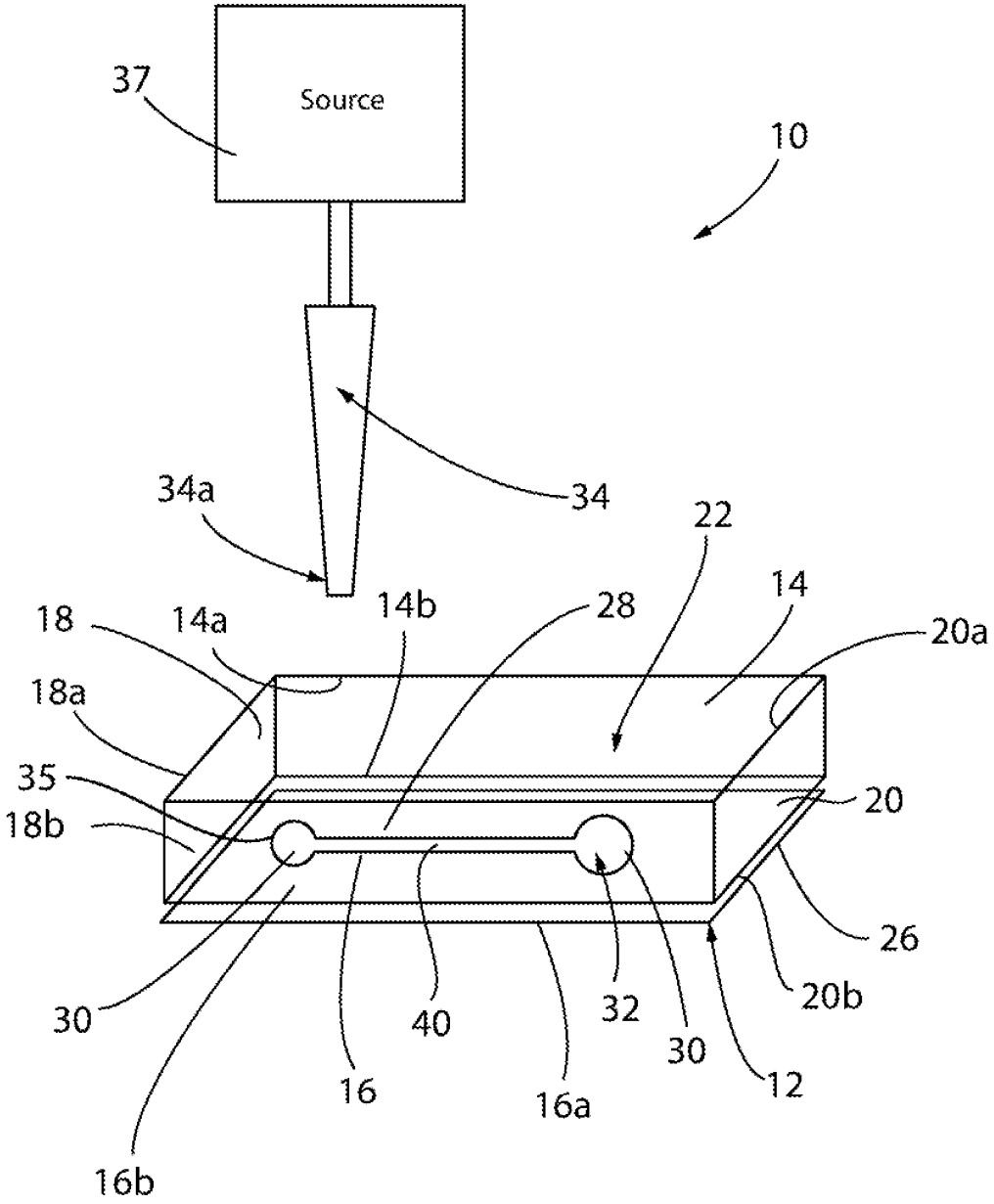
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Source
10
34a
34
22
14
18
14a
14b
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20a
18a
35
20
18b
26
30
40
32
30
20b
12
16
16a
16b

FIG. 1

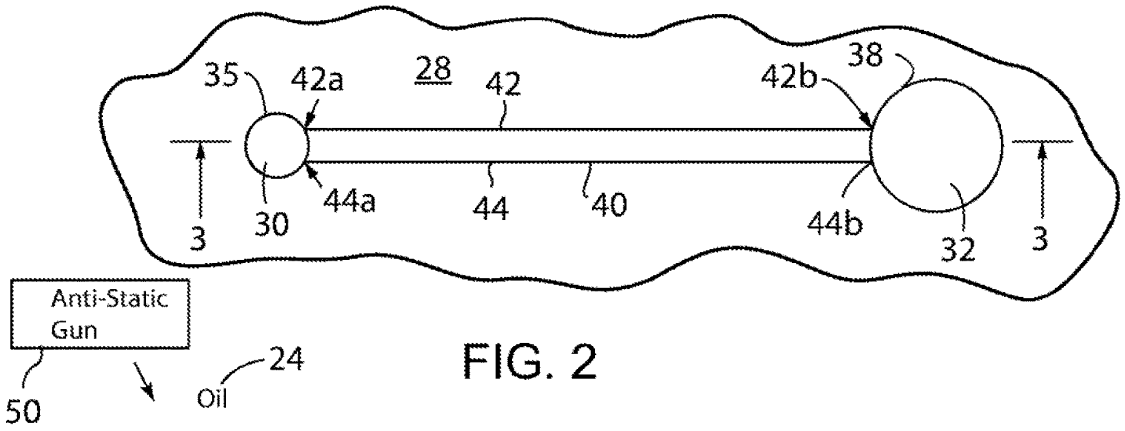

FIG. 2

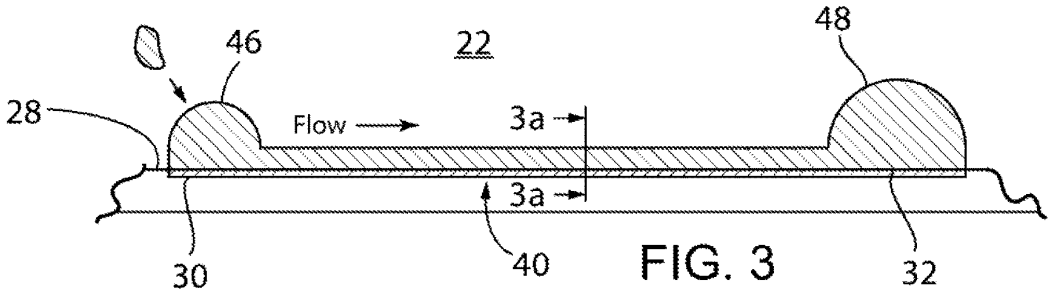

FIG. 3

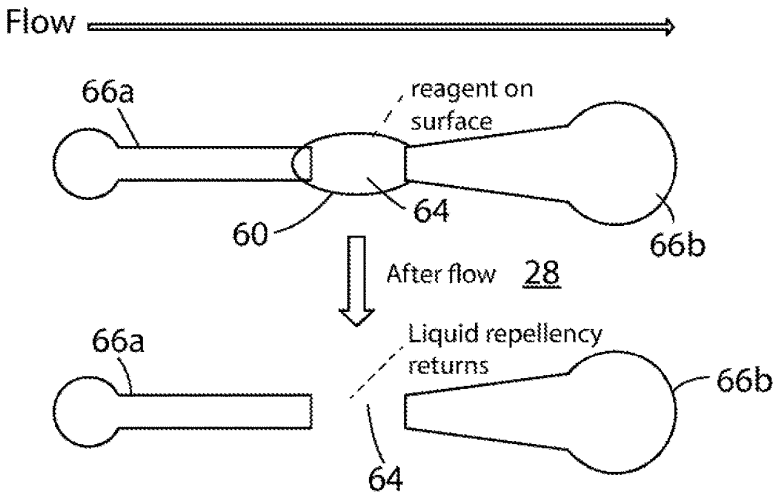

FIG. 4

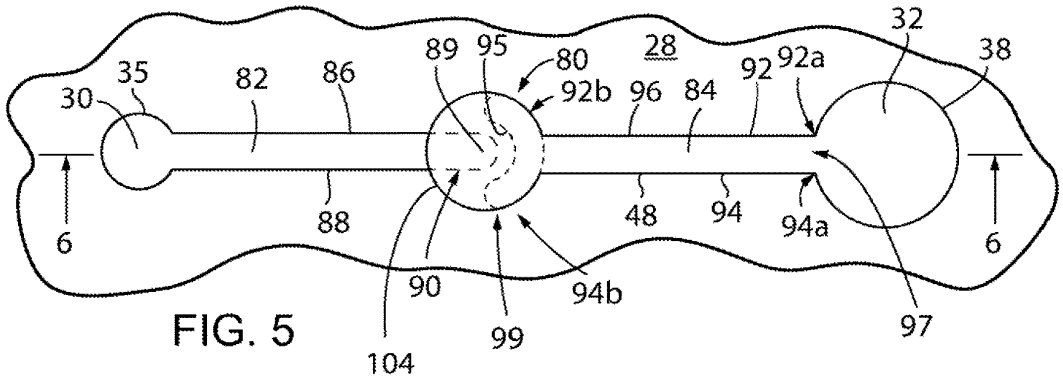

FIG. 5

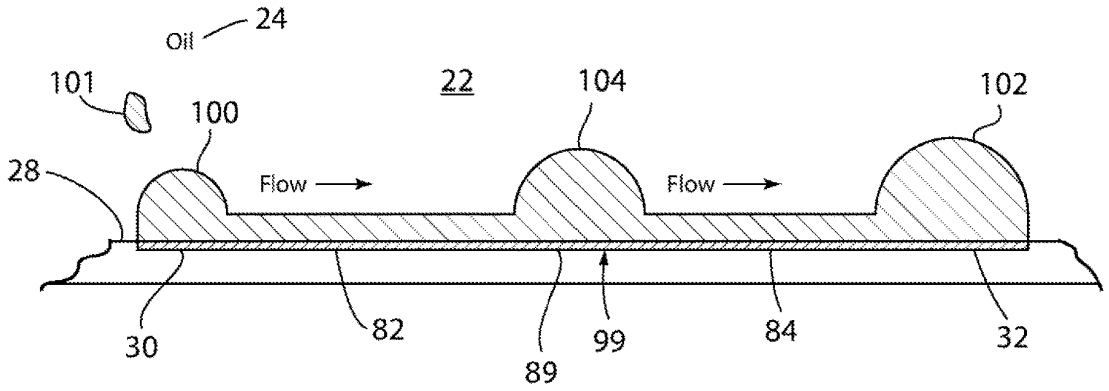

FIG. 6

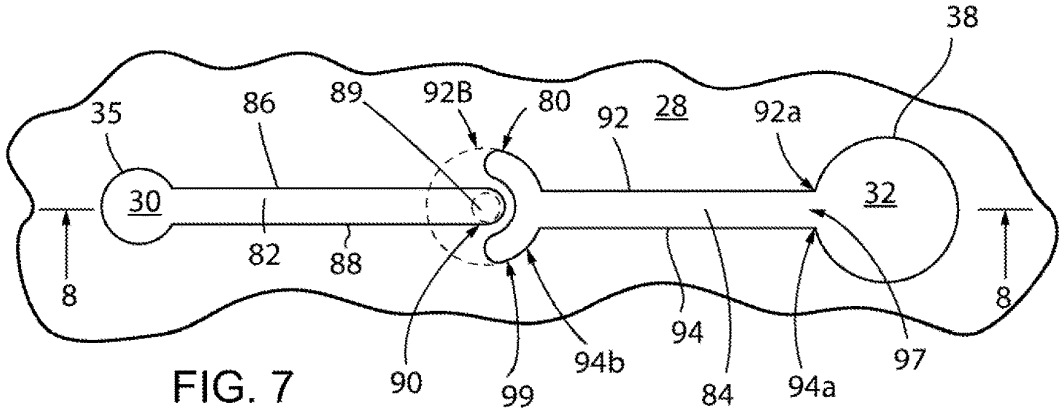

FIG. 7

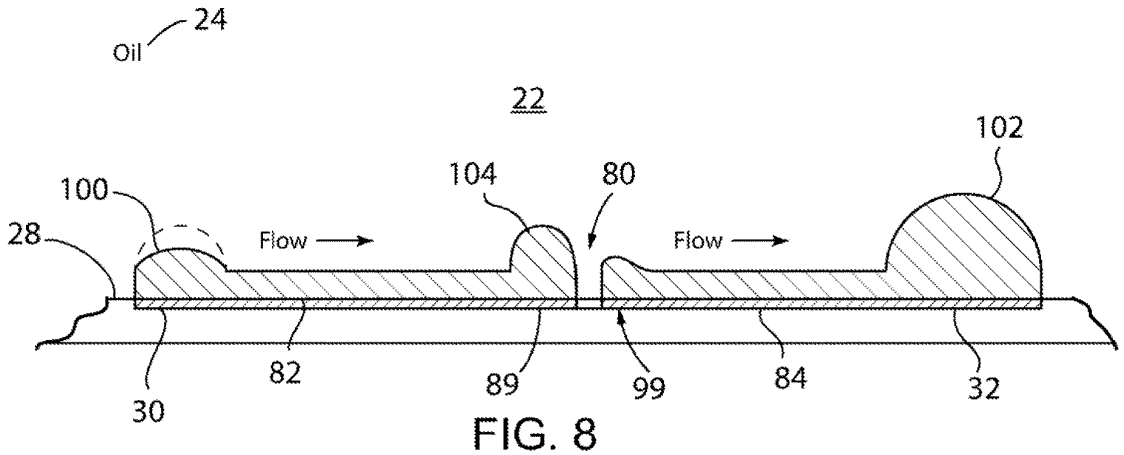

FIG. 8

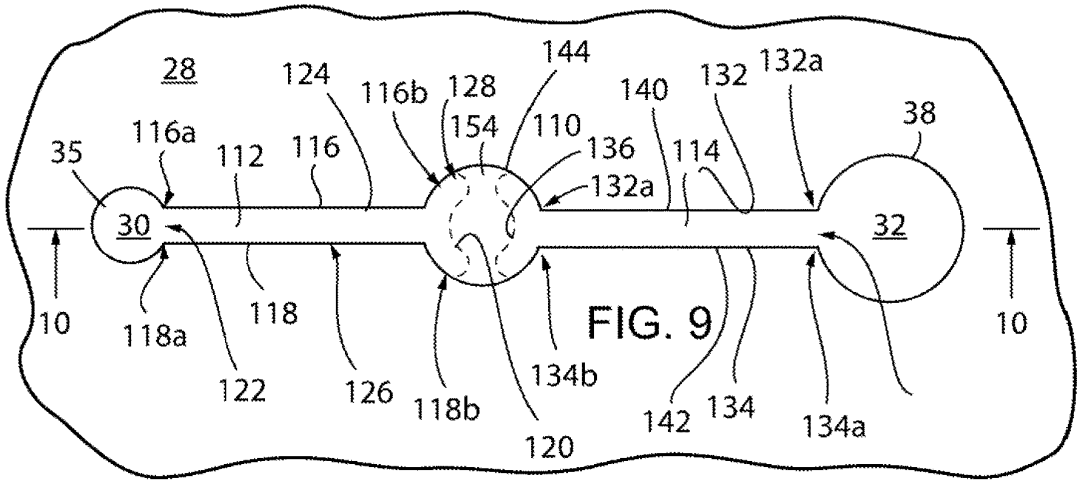

FIG. 9

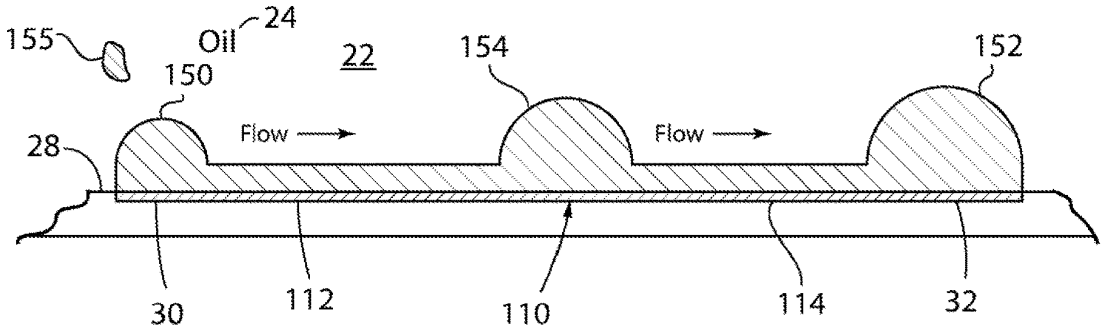

FIG. 10

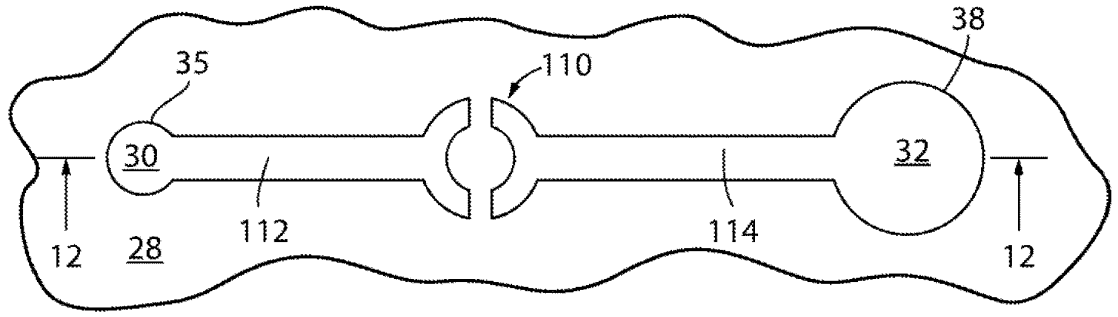

FIG. 11

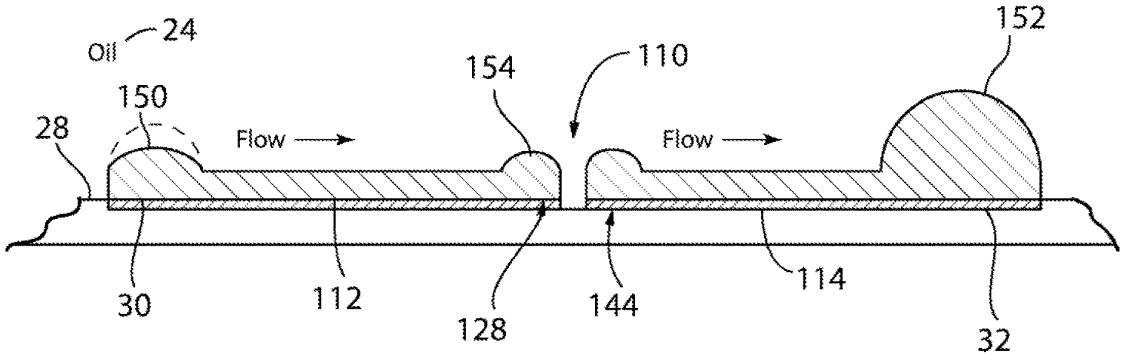

FIG. 12

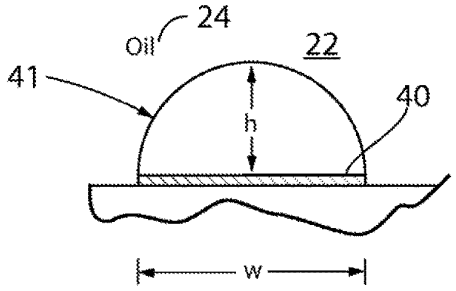

FIG. 3a

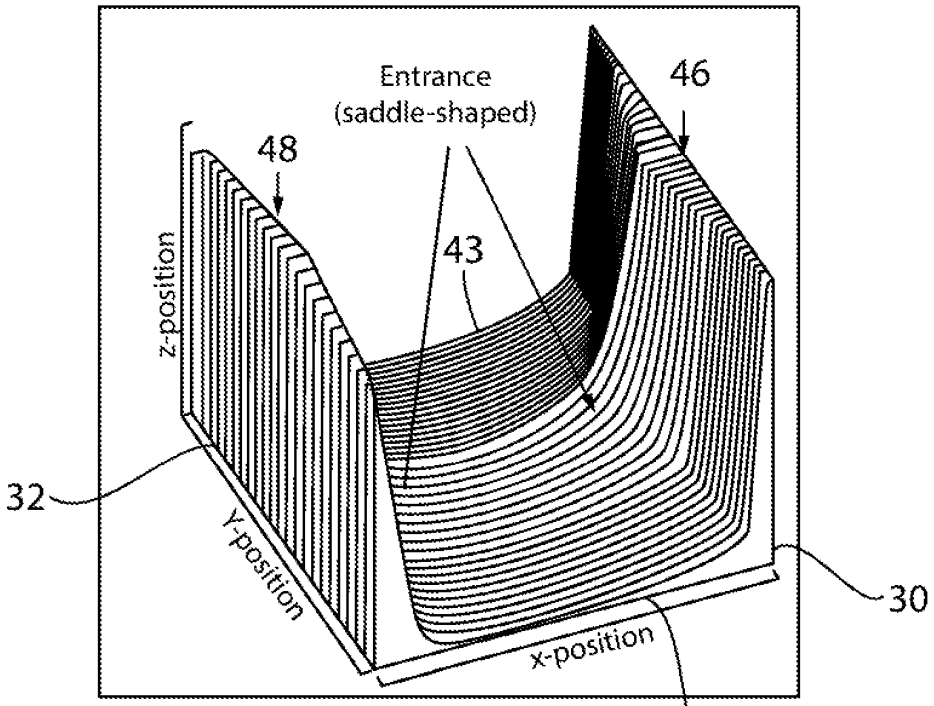
Entrance
(saddle-shaped)
46
48
43
z-position
32
Y-position
x-position
30
41

FIG. 13

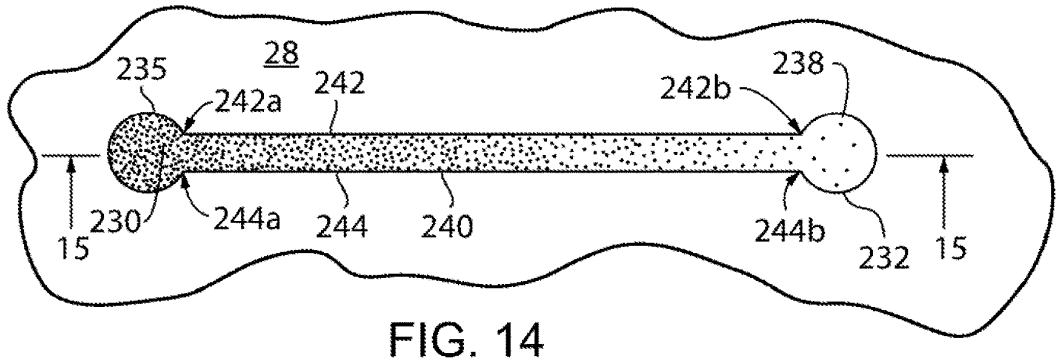
28
235
242a
242
242b
238
15
230
244a
244
240
244b
232
15

FIG. 14

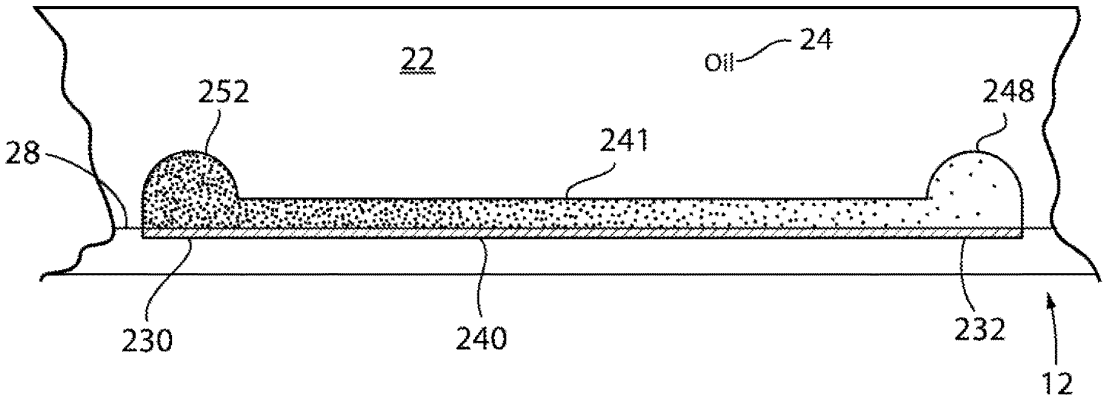
22
Oil
24
252
248
28
241
230
240
232
12

FIG. 15

OPEN MICROFLUIDIC SYSTEM AND VARIOUS FUNCTIONAL ARRANGEMENTS THEREFORE

CROSS-REFERENCE TO RELATED APPLICATION

This application claims benefit from U.S. Provisional Application Ser. No. 62/868,378, filed Jun. 28, 2019, the entirety of which is incorporated herein.

REFERENCE TO GOVERNMENT GRANT

This invention was made with government support under CA181648 awarded by the National Institutes of Health and W81XWH-16-1-0514 awarded by the ARMY/ARO. The government has certain rights in the invention.

FIELD OF THE INVENTION

This invention related generally to microfluidics, and in particular, to an open microfluidic system including the extreme wettability of exclusive liquid repellency (ELR), open microchannels with high lateral resolution and low profile and with various valve arrangements, capable of a broad range flow rates, and capable of spatially and temporally trapping particles.

BACKGROUND AND SUMMARY OF THE INVENTION

Open microfluidics has been defined as a microfluidic system with at least one solid boundary confining the fluid removed, exposing the fluid either to air (i.e., single-liquid-phase) or a second fluid (i.e., multi-liquid-phase). One disadvantage of single-liquid-phase open systems is their sensitivity to evaporation. To overcome this limitation many open microfluidic systems employ an oil overlay (similar to the oil-overlaid microdroplets used for decades for the in vitro study of early embryo development) to prevent detrimental fluid loss via evaporation and airborne contamination through the liquid-air interface. Important advantages of open microfluidics include accessibility, bubble elimination and ease of use. The liquid/air or liquid/liquid interface above and surrounding the channel provides direct physical access to the fluid of interest, e.g., enabling localized interrogation of cellular samples with their biophysics or biochemistry. Also, without the need to bond to another surface, open microfluidic devices are generally easy-to-make and easy-to-use (e.g., elimination of bubble trapping and associated device failures), reducing the adoption barrier to end users.

Under oil open microfluidics has been limited in its functional/operational range due to the lack of lateral flow. Recently, lateral flow was introduced to under oil open microfluidics to expand its functionality. Fundamental to most microfluidic systems is their ability to control mass transport (e.g., maintaining steady flow, varying flow rate, and turning flow on and off). However, reported under oil open-channel systems exhibit a limited flow range (from both the upper and lower limits) thus limiting potential applications. For example, the maximum flow rate is several orders of magnitude lower than the typical range for closed channels. The reported techniques form channels via a two-step process whereby the channels are initially filled in air and then subsequently overlaid with oil. This two-step process presents a number of technical challenges that limit the scope of their geometries and application. From a practical operational perspective, the initial in-air step (prior to the introduction of an oil overlay) restricts the channel width to the millimeter scale due to volume loss via evaporation. Millimeter scale channels are limited in their ability to spatially and temporally organize cellular samples (e.g., mammalian and bacterial cells are less than 10 μm), and flexibility in control of mass transport. In addition to extending the range of flow rates, open microchannels should also be able to provide the ability to turn flow on and off.

Therefore, it is a primary object and feature of the present invention to provide an open microfluidic system that allows for increased accessibility, minimized evaporation and airborne contamination, and ease of use over prior systems.

It is a further object and feature of the present invention to provide an open microfluidic system that allows for a high lateral resolution of fluidic channels (e.g., a few microns in channel width, spacing, and height).

It is a still further object and feature of the present invention to provide an open microfluidic system that allows for confinement of various cellular samples (e.g., mammalian cells, bacteria, and fungi) in open fluid.

It is a still further object and feature of the present invention to provide an open microfluidic system that enables flow control covering several orders of magnitude.

It is a still further object and feature of the present invention to provide an open microfluidic system that includes various valve arrangements with the ability to reversibly turn fluid flow through the system on and off.

It is a still further object and feature of the present invention to provide an open microfluidic system including a single-use valve for use in a sample loading for streamlined multi-step assays.

It is a still further object and feature of the present invention to provide an open microfluidic system that is simple to utilize and inexpensive to manufacture.

In accordance with the present invention, an open microfluidic system is provided. The open microfluidic system includes a microfluidic device having a reservoir adapted for receiving an oil therein. The reservoir is defined by a surface configured to repel an aqueous solution. A hydrophilic input and a hydrophilic output are patterned on the surface. The output is spaced from the input. A hydrophilic strip interconnects the input and the output.

The strip includes a first channel having a first end connected to the input, a second channel having a first end connected to the output, and a valve configured to selectively fluidically connect the second ends of the first and second channel. The valve includes a second end of the first channel and a second end of the second channel. The valve may have one of plurality of configurations. By way of example, the valve may include a dried reagent fluidically interconnecting the second end of the first channel and the second end of the second channel. Fluid flowing over the dried reagent picks-up and re-dissolves the dried reagent therein, thereby exposing a portion of the surface between the first and second hydrophilic channels and fluidically isolates the first channel from the second channel.

Alternatively, the second end of the second channel may have a horseshoe configuration. In addition, the second end of the first channel has a horseshoe configuration. A droplet having a first dimension may be deposited on the surface. When the droplet communicates with the second end of the first channel and the second end of the second channel, the valve is closed. When the droplet has a second dimension,

the droplet fluidically isolates the second end of the first channel form the second end of the second channel, thereby opening the valve.

In accordance with a further aspect of the present invention, a method of fabricating an open microfluidic system is provided. The method includes the step of providing a microfluidic device including a reservoir defined by a surface configured to repel an aqueous solution. A hydrophilic input, a hydrophilic output and a hydrophilic strip interconnecting the input and output are patterned on the surface. The reservoir is filled with an oil. An input droplet of the aqueous solution is positioned on the input and an output droplet of the aqueous solution is positioned on the output. The input droplet and the output droplet are fluidically connected along the strip with the aqueous solution.

The step of fluidically connecting the input droplet and the output droplet along the strip includes the step of generating an external perturbation on the oil in the reservoir. The external perturbation on the oil may be generated by an anti-static gun Which repetitively pumps ionized air at the oil. The strip may include a valve, the valve having a first open configuration fluidically isolating the input from the output and a second closed configuration wherein the input and the output are in fluidic communication. The strip includes a first channel having a first end connected to the input and a second channel having a first end connected to the output. The valve is configured to selectively fluidically connect the first and second channels. The valve may include a second end of the first channel and a second end of the second channel. A dried reagent may fluidically interconnecting the second end of the first channel and the second end of the second channel. A fluid flowing over the dried reagent picks-up and re-dissolves the dried reagent therein so as to expose a portion of the surface between the first and second hydrophilic channels, thereby opening the valve.

Alternatively, the second end of the second channel may have a horseshoe configuration and the second end of the first channel may have a horseshoe configuration. The valve includes a droplet having a first dimension wherein the droplet communicates with the second end of the first channel and the second end of the second channel thereby closing the valve and a second dimension wherein the droplet fluidically isolates the second end of the first channel form the second end of the second channel thereby opening the valve.

In accordance with a still further aspect of the present invention, a single-use valve is provided. The valve includes a microfluidic device having a reservoir defined by a surface configured to repel an aqueous solution. The reservoir is configured for receiving oil therein. First and second hydrophilic channels are patterned on surface. The first and second hydrophilic channels spaced from each other. A dried reagent interconnects the first and second hydrophilic channels. Fluid flowing over the dried reagent picks-up and re-dissolves the dried reagent therein, thereby exposing a portion of the surface between the first and second hydrophilic channels and fluidically isolating the first hydrophilic channel from the second hydrophilic channel.

BRIEF DESCRIPTION OF THE DRAWINGS

The drawings furnished herewith illustrate a preferred methodology of the present invention in which the above advantages and features are clearly disclosed as well as others which will be readily understood from the following description of the illustrated embodiment.

In the drawings:

FIG. **1** is a schematic view of an open microfluidic system in accordance with the present invention;

FIG. **2** is a top plan view showing a portion of the open microfluidic system of FIG. **1**;

FIG. **3** is a cross-sectional view of the open microfluidic system of the present invention taken along line **3-3** of FIG. **1**;

FIG. **3***a* is a cross-sectional view of the open microfluidic system of the present invention taken along line **3***a*-**3***a* of FIG. **3**;

FIG. **4** is a schematic, top plan view of the open microfluidic system of FIG. **1** including a single use valve shown in a first connected position and a second disconnected position;

FIG. **5** is a top plan view showing a portion of the open microfluidic system of FIG. **1** including an alternate valve shown in a first connected position;

FIG. **6** is a cross-sectional view of the open microfluidic system of the present invention taken along line **6-6** of FIG. **5**;

FIG. **7** is a top plan view showing a portion of the open microfluidic system of FIG. **5** including the valve shown in a second disconnected position;

FIG. **8** is a cross-sectional view of the open microfluidic system of the present invention taken along line **8-8** of FIG. **7**;

FIG. **9** is a top plan view showing a portion of the open microfluidic system of FIG. **1** including a further alternate valve shown in a first connected position;

FIG. **10** is a cross-sectional view of the open microfluidic system of the present invention taken along line **10-10** of FIG. **9**;

FIG. **11** is a top plan view showing a portion of the open microfluidic system of FIG. **9** including the valve shown in a second disconnected position;

FIG. **12** is a cross-sectional view of the open microfluidic system of the present invention taken along line **12-12** of FIG. **11**;

FIG. **13** is a schematic, isometric view of the open microfluidic system of the present invention;

FIG. **14** is a top plan view showing a portion of the open microfluidic system of FIG. **1** utilized to generate an under-oil gradient; and

FIG. **15** is a cross-sectional view of the open microfluidic system of the present invention taken along line **15-15** of FIG. **14**.

DETAILED DESCRIPTION OF THE DRAWINGS

ELR is a phenomenon observed in solid-liquid-liquid three phase systems, where a solid surface shows complete repellency to a liquid (with a contact angle (CA)=180°) when exposed to a second liquid. This phenomenon is observed when a particular thermodynamic boundary condition is satisfied, for example, by the equation:

$$\gamma_{S/Lcp}+\gamma_{Ldp/Lcp}\leq\gamma_{S/Ldp} \quad \text{Equation (1)}$$

wherein: γ is the interfacial tension; S is solid; Lcp is a liquid of continuous phase; and Ldp is a liquid of dispersed phase. ELR enables additional fluidic control, robust on-chip cell culture, and improved processing of rare cell samples in open aqueous fluid under oil.

In systems employing double-ELR, there is selective and complete repellency of two immiscible liquids from adjacent surfaces through surface chemistry patterning. Fluids are naturally contained on "preferred" surfaces of having a

lower contact angle, which lowers free energy of the system. The boundary between surface patterns thus creates a virtual barrier (i.e., an energetic impediment) to fluid expansion from its footprint (i.e., the contact area between the fluid and its preferred surface). Double-ELR offers the theoretical maximum virtual barrier to both aqueous fluid and oil with a CA of 180° on their "non-preferred" surfaces (i.e., oil on glass, media on PDMS), and thus robustly confining the fluids to their preferred surfaces (i.e., oil on PDMS, media on glass). This virtual barrier is important to stabilize the three phase contact line. If fluid spreads from its original footprint, it is completely repelled by the non-preferred surface and recedes to the original pattern when the system is allowed to equilibrate. Double-ELR allows for spontaneous, uncompromised oil/media separation (without the need for surfactant), and thus, under oil sweep in open microfluidic designs. Under oil sweep is accomplished by simply dragging media across the patterned surface under oil, resulting in a specific volume of media (with or without cells) being spontaneously dispensed onto the patterned areas (i.e., microchannels or spots) and leaving the background clean with minimized sample loss and device fouling. As hereinafter described, by exploiting double-ELR and the technique of under oil sweep, open microchannels under oil are achieved with improved lateral resolution (for example, ~30 μm in both width and spacing), low profiles (for example, ~1 μm in height) capable of cell trapping, convective bulk flow covering eight orders of magnitude (for example, from 6 mL/min to 13 pL/min), and fully reversible fluidic valves.

Referring to FIGS. **1-3**, a schematic drawing depicting a system for carrying out a first aspect of the methodology of the present invention is generally designated by the reference numeral **10**. System **10** includes microfluidic device **12** defined by first and second generally parallel, spaced side walls **14** and **16** respectively, interconnected by first and second generally parallel, spaced end walls **18** and **20**, respectively. First and second side walls **14** and **16** respectively, and first and second end walls **18** and **20** respectively, define reservoir **22** for receiving a fluid, such as oil **24** or like, for reasons hereinafter described. It can be understood that oil **24** may be any liquid showing limited miscibility with water or any aqueous media. Upper edges **14***a* and **16***a* of first and second side walls **14** and **16** respectively, and upper edges **18***a* and **20***a* of end walls **18** and **20**, respectively, define an opening **26** for allowing access to reservoir **22**. Lower edges **14***b* and **16***b* of first and second side walls **14** and **16** respectively, and lower edges **18***b* and **20***b* of end walls **18** and **20**, respectively, are interconnected by an exclusive liquid repellency (ELR) surface **28** which communicates with reservoir **22**. ELR surface **28** is a hydrophobic solid surface having specific surface chemical and physical conditions intended to repel aqueous solutions, as hereinafter described. It can appreciated that while microfluidic device **10** has a generally rectangular, box-like configuration, other configurations are possible without deviating from the scope of the present invention.

In the depicted embodiment, surface patterning is provided on ELR surface **28** of open microfluidic system **10** of the present invention. For example, surface patterning of ELR surface **28** may be done on a PDMS-grafted glass substrate using a reusable PDMS stamp and O2 plasma diffusion treatment. More specifically, a hydrophilic input spot **30** is pre-patterned on ELR surface **28** of microfluidic device **12** at a selected first location and a hydrophilic output spot **32** is pre-patterned on ELR surface **28** of microfluidic device **12** at a selected second location, axially spaced from the first location. Input spot **30** has an outer periphery intersecting ELR surface **28** at boundary **35**. Similarly, output spot **32** has an outer periphery intersecting ELR surface **28** at boundary **38**. It is contemplated for output spot **32** to have a greater cross-sectional area than input spot **30**, for reasons hereinafter described. However, input spot **30** and output spot **32** may be other configurations without deviating from the scope of the present invention.

Input spot **30** and output spot **32** are interconnected by a hydrophilic strip **40** pre-patterned on and extending axially along ELR surface **28**. Strip **40** is defined by first and second, generally parallel edges **42** and **44**, respectively, which intersect ELR surface **28**. First and second edges **42** and **44**, respectively, have corresponding first ends **42***a* and **44***a*, respectively, which intersect boundary **35** of input spot **32** and corresponding second ends **42***b* and **44***b*, respectively, which intersect boundary **38** of output spot **32**. The functionality of microfluidics leverages channel dimensions of tens to hundreds of microns. By way of example, strip **40** has width ranging from 10 to 200 micrometers (μm). It is further contemplated for the surface patterning on ELR surface **28** to have other configurations. For example, strip **40** may be wider at second ends **42***b* and **44***b* and narrower at first ends **42***a* and **44***a* to promote passive pumping of an aqueous media in system **10**, hereinafter described. In addition, it is contemplated to provide input and output spots **30** and **32**, respectively, at different locations along strip **40**, without deviating from the scope of the present invention.

In operation, reservoir **22** of microfluidic device **12** is filled with a selected fluid, such as oil **24**. After flooding the reservoir **22** with oil **24**, one of more injectors **34** are configured to deliver input droplet **46** of a desired aqueous media on input spot **30** and outlet droplet **48** of a desired aqueous media on output spot **32**. In order to generate flow between input spot **30** and output spot **32** along strip **40**, it can be understood that the aqueous media must displace a thin layer of oil **24** at the interface of oil **24** and strip **40** come into contact with strip **40**. To overcome the energy barrier for the displacement of oil **24** and facilitate the flow of the aqueous media along strip **40**, it is contemplated to provide an external perturbation on the interface of oil **24** and strip **40**, e.g., utilizing anti-static gun **50**. Other mechanisms such as an on-chip micro transducer, a surface acoustic wave generator, and/or paramagnetic beads moved with a magnet may be used to provide the external perturbation on the interface of oil **24** and strip **40**, without deviating scope of the present invention.

To generate the external perturbation on the interface of oil **24** and strip **40**, anti-static gun **50** is positioned a selected distance, e.g., 5 centimeters, above the upper surface of oil **24**. Anti-static gun **50** repetitively pumps streams of ionized air at oil **24** so as to provide alternate positive and negative charges on oil **24**. Oil **24** vibrates in response to the repetitive pumping of the ionized air by anti-static gun **50**, thus causing a momentum to be applied to the interface of oil **24** and strip **40**. This momentum helps the aqueous media overcome the energy barrier for the displacement of oil **24** and allow for the aqueous media to flow along strip **40** to interconnect input spot **30** and output spot **32**. The aqueous media along strip **40** defines a microchannel **41** having a height h, a width w and a length, FIG. **3***a*.

In order to generate fluid flow from input spot **30** to output spot **32**, injector **34**, operatively connected to an aqueous media source **37**, may be used to deliver aqueous media to input droplet **46** such that the fluidic pressure in input droplet **46** urges the aqueous media along strip **40** toward output droplet **48**. Alternatively, because input droplet **46** has a

smaller radius of curvature than output droplet **48**, a larger pressure exists on input spot **30**. The resulting pressure gradient causes aqueous media to flow from input droplet **46**, along strip **40**, towards output droplet **48**. It can be understood that by sequentially depositing additional drops of aqueous media to input droplet **46** on input spot **30** with injector **34**, the resulting pressure gradient will cause the aqueous media to flow along strip **40** towards output droplet **48** on output spot **32**. As a result, fluid flows along strip **40** from input droplet **46** to output droplet **48**. It can be understood that in addition to generating the flow of aqueous media in system **10** utilizing passive pumping, as heretofore described, other mechanisms (e.g., a syringe pump) may be used without deviating from the scope of the present invention.

It has been determined that microchannel **41** has a similar height/width (h/w) ratio (e.g., approximately 1:13), which is independent of the width of strip **40**. Hence, it can be appreciate that cellular samples flowing in microchannel **41** may be confined within a designated area simply by adjusting a dimension (i.e., either the width or the height) of portion **43** of microchannel **41** to be comparable or smaller than the objects being confined (e.g., a single cell). The small height/width ratio of the open microchannels means confinement of cellular samples can be achieved with microchannels having relatively large widths. In contrast to closed systems, the geometry and effect of surface tension in open systems result in a gradual rather than abrupt channel entrance. This produces a saddle-shaped geometry at the connection of microchannel **41** to input and output spots **30** and **32**, respectively, FIG. **13**. Due to the unique entrance geometry, cells entering microchannel **41** follow a contour line whose height is comparable to the size of a single cell, named an entrance plume. The entrance plume can affect the constraint of cellular samples in adjacent spots. If the length of a channel is less than two times the length of the entrance plume, then the height at the center of the channel is enough for cells to flow from input spot **30** to output spot **32**.

Critical to the function of microfluidic systems is the ability to have flow across multiple scales. To extend the function of open microfluidic systems, it is necessary that the flow rates at both the upper and lower limits (i.e., for maximum and minimum flow respectively) be expanded. Flow is important for many applications in biomedical research where control of mass transport (e.g., a constant drug delivery rate) and/or mechanical cues (e.g., stable shear force on cell-surface adhesion) is necessary. The volumetric flow rate (Q) of fluid in a channel may be calculated according to the expression:

$$\Delta P = R_h Q \qquad \text{Equation (2)}$$

wherein: ΔP is the pressure drop and R_h is the hydrodynamic resistance.

Given the lack of physical walls in open microfluidic system **10**, robust confinement of fluid in open channels can be challenging, especially when high ΔP is applied. To increase the upper limit of flow rate in open-channel designs, a maximum virtual barrier to the fluid, such as that enabled by double-ELR, is desired. On the other end, to establish a low flow rate, a sufficiently high R_h is necessary to limit convective bulk flow. It can be understood that R_h can be controlled by varying the dimensions of microchannel, either by making microchannel **41** longer or by reducing the cross sectional area thereof. More specifically, for a given resistance, the length of microchannel **41** must be increased exponentially to offset the change in the cross sectional area of microchannel **41**. Hence, to establish a low flow rate across a short distance (e.g., a few hundred microns, a typical distance in which capillary exchange occurs in vivo), a reduced cross sectional area is necessary. To maintain $\Delta_{critical}$, and thus a steady flow through microchannel **41**, a syringe pump may be utilized to add volume of aqueous media to inlet droplet **46**, while simultaneously removing the same volume of aqueous media from outlet droplet **48**.

Referring to FIG. **4**, it is contemplated for system **10** to include a dissolvable single-use smart "valve." By way of example, strip **40** may be defined by first and second hydrophilic channels **66***a* and **66***b* patterned on ELR surface **28** of microfluidic device **12**. With reservoir **22** dry and free of fluids, reagent **60** of interest in solution is deposited onto ELR surface **28** at a location **64** interconnecting first and second hydrophilic channels **66***a* and **66***b*, respectively. Reagent **60** is allowed to dry and physically adsorb onto surface **28**. Once reagent **60** is dried on surface **28**, reservoir **22** of microfluidic device **12** is filled with a selected fluid, such as oil **24**.

With reservoir **22** filled with oil, an aqueous solution of interest may be flowed from first hydrophilic channel **66***a*, over reagent **60** at location **64**, to second hydrophilic channel **66***b*, as heretofore described. It can be understood that as the aqueous solution of interest flows from first hydrophilic channel **66***a*, over reagent **60** at location **64**, to second hydrophilic channel **66***b*, the aqueous solution of interest picks-up and re-dissolves the desiccated reagent **60** therein so as to carry reagent **60** to second hydrophilic channel **66***b*. Once all the available reagent **60** is dissolved, ELR surface **28** is returned back to a liquid repellent state, thus disconnecting first hydrophilic channel **66***a* from second hydrophilic channel **66***b*. This arrangement allows for reagent **60** at location **64** to act as a valve, serving as both a reagent delivery device and an autonomous self-regulating timer that shuts off liquid flow once all reagent **60** is delivered to second hydrophilic channel **66***b*.

Alternatively, it is contemplated to incorporate a reversible valve into system **10**. It can be appreciated that open channels present a unique challenge in the design of reversible valves due to the lack of physical walls whereby a mechanism capable of connecting, disconnecting and reconnecting fluid flow can be easily deployed.

Referring to FIGS. **5-8**, an alternate embodiment of a valve is generally designated by the reference numeral **80**. By way of example strip **40** may be defined by first and second hydrophilic channels **82** and **84** patterned on ELR surface **28** of microfluidic device **12**. First channel **82** extends along an axis and is defined by first and second, generally parallel edges **86** and **88**, respectively, which intersect ELR surface **28**. First and second edges **86** and **88**, respectively, have first ends which intersect boundary **35** of input spot **30** and corresponding second ends which intersect each other and define hydrophilic spot **89** at terminal end **90** of first channel **82**. Second channel **84** extends along an axis and is defined by first and second edges **92** and **94**, respectively, and concave edge **95** which intersect ELR surface **28**. More specifically, first and second edges **92** and **94** respectively, include first ends **92***a* and **94***a*, respectively, which define output end **97** of second channel **84** and intersect boundary **38** of output spot **32**. Parallel portions **96** and **98** of first and second edges **92** and **94**, respectively, extend from first ends **92***a* and **94***a* and intersect second ends **92***b* and **94***b* of first and second edges **92** and **94**, respectively. Second ends **92***b* and **94***b* of first and second edges **92** and **94**, respectively, diverge from each other and extend on opposite sides of terminal end **90** of first channel **82**. Second ends **92***b* and **94***b* of first and second edges **92** and **94**

respectively, are interconnected by concave edge **95** which extends about terminal end **90** of first channel **82**. Second ends **92***b* and **94***b* of first and second edges **92** and **94**, respectively, and concave edge **95** define a generally horseshoe-shaped input end **99** of second channel **84**.

In operation, reservoir **22** of microfluidic device **12** is filled with a selected fluid, such as oil **24**. One of more injectors **34** are configured to deliver input droplet **100** of a desired aqueous media on input spot **30**, outlet droplet **102** of a desired aqueous media on output spot **32**, and bridge droplet **104** on spot **89** at terminal end **90** of first channel **82**. It is intended for bridge droplet **104** to be of sufficient dimension to overlap and communicate with input end **99** of second channel **84** so as to fluidically connected first and second channels **82** and **84**, respectively, FIGS. **5-6**. It is intended for bridge droplet **104** to fluidically connect first and second channels **82** and **84**, respectively, thereby closing valve **80** and allowing for fluid flow therebetween.

In order to generate flow between input spot **30** and output spot **32** along strip **40**, it is contemplated to provide an external perturbation on the interface of oil **24** and strip **40** utilizing anti-static gun **50**. As heretofore described, oil **24** vibrates in response to the repetitive pumping of the ionized air by anti-static gun **50**, thus causing a momentum to be applied to the interface of oil **24** and strip **40**. This momentum helps the aqueous media overcome the energy barrier for the displacement of oil **24** and allow for the aqueous media to flow along strip **40** to fluidically connect input droplet **100** on input spot **30** to bridge droplet **104** and to fluidically connect bridge droplet **104** and output droplet **102** on output spot **32**. Because input droplet **100** has a smaller radius of curvature than bridge droplet **104**, a larger pressure exists on input spot **30**. The resulting pressure gradient causes aqueous media to flow from input droplet **100**, along first channel **82**, towards bridge droplet **104**. Similarly, because bridge droplet **104** has a smaller radius of curvature than output droplet **102**, a larger pressure exists on spot **89**. The resulting pressure gradient causes aqueous media to flow from bridge droplet **104** along second channel **84**, towards output droplet **102**. It can be understood that by sequentially depositing additional drops **101** of aqueous media to input droplet **100** on input spot **30** with injector **34**, the resulting pressure gradient will cause the aqueous media to flow along first channel **82**, through bridge droplet **104**, along second channel **84** towards output droplet **102** on output spot **32**. As a result, fluid flows along strip **40** from input droplet **100** to output droplet **102**, FIG. **6**.

By terminating the depositing of drops **101** of aqueous media, it can be understood that the volume of aqueous media flowing in bridge droplet **104** decreases, while the aqueous media continues to output droplet **102**, thereby reducing the dimension of bridge droplet **104**, FIGS. **7-8**. More specifically, as the dimension abridge droplet **104** is reduced, the volume in bridge droplet **104** reaches a critical point (i.e., the minimum amount of liquid required to maintain the fluidic connection between first and second channels **82** and **84**, respectively (hereinafter referred to as "$V_{critical}$"). Once bridge droplet **104** reaches $V_{critical}$, termination end **90** of first channel **82** becomes isolated from input end **99** of second channel **84**, thereby opening or disconnecting valve **80** and terminating the fluid flow between input spot **30** and output spot **32** along strip **40**. Reinstitution of fluid flow between input spot **30** and output spot **32** along strip **40** may be achieved by simply adding aqueous media to bridge droplet **104** to reestablish a fluidic connection between first and second channels **82** and **84**, respectively, and sequentially depositing additional drops **101** of aqueous media on input droplet **100**.

It can be understood that $V_{critical}$ is dependent upon the geometry and the size of the portion of ELR surface **28** between termination end **90** of first channel **82** and input end **99** of second channel **84** (hereinafter referred to as the ELR gap). A larger ELR gap requires a larger $V_{critical}$ to maintain the connection between first and second channels **82** and **84**, respectively. The connection time ($\Delta t_{connection}$) of valve **80** is defined as the time to reduce the initial volume of bridge droplet **104** ($V_{initial}$) to $V_{critical}$. It can be appreciated that a larger $V_{critical}$ (e.g., a valve with a larger ELR gap) results in a shortened $\Delta t_{connection}$ for a given Q.

It is noted that fluid flow through valve **80** may be reversed by providing outlet droplet **102** with a smaller radius of curvature than bridge droplet **104** and by providing bridge droplet **104** with a smaller radius of curvature than input droplet **100**. The resulting pressure gradient causes aqueous media to flow from output droplet **104**, along second channel **84**, towards bridge droplet **104**. Similarly, because bridge droplet **104** has a smaller radius of curvature than input droplet **100**, the resulting pressure gradient causes aqueous media to flow from bridge droplet **104**, along first channel **82**, towards input droplet **100**. It can be understood that by sequentially depositing additional drops **101** of aqueous media to outlet droplet **102**, the resulting pressure gradient will cause the aqueous media to flow along second channel **84**, through bridge droplet **104**, along first channel **82** towards input droplet **100**. As a result, fluid flows along strip **40** from output droplet **102** to input droplet **100**.

Referring to FIGS. **9-12**, a still further embodiment of a valve is generally designated by the reference numeral **110**. By way of example, strip **40** may be defined by first and second hydrophilic channels **1112** and **114** patterned on ELR surface **28** of microfluidic device **12**. First channel **112** extends along an axis and is defined by first and second edges **116** and **118**, respectively, and concave edge **120** which intersect ELR surface **28**. More specifically, first and second edges **116** and **118**, respectively, include first ends **116***a* and **118***a*, respectively, which define input end **122** of first channel **112** and intersect boundary **35** of input spot **30**. Parallel portions **124** and **126** of first and second edges **116** and **118**, respectively, extend from first ends **116***a* and **118***a* and intersect second ends **116***b* and **118***b* thereof. Second ends **116***b* and **118***b* of first and second edges **116** and **118**, respectively, diverge from each other. Second ends **116***b* and **118***b* of first and second edges **116** and **118**, respectively, are interconnected by concave edge **120**. Second ends **116***b* and **118***b* of first and second edges **116** and **118**, respectively, and concave edge **120** define a generally horseshoe-shaped output end **128** of first channel **112**.

Second channel **114** extends along an axis and is defined by first and second edges **132** and **134**, respectively, and concave edge **136** which intersect ELR surface **28**. More specifically, first and second edges **132** and **134**, respectively, include first ends **132***a* and **134***a*, respectively, which define output end **138** of second channel **114** and intersect boundary **38** of output spot **32**. Parallel portions **140** and **142** of first and second edges **132** and **134**, respectively, extend from first ends **132***a* and **134***a* and intersect second ends **1132***b* and **134***b* of first and second edges **1132** and **134**, respectively. Second ends **132***b* and **134***b* of first and second edges **132** and **134**, respectively, diverge from each other. Second ends **132***b* and **134***b* of first and second edges **132** and **134**, respectively, are interconnected by concave edge **136**. Second ends **132***b* and **134***b* of first and second edges **132** and **134**, respectively, and concave edge **136** define a

generally horseshoe-shaped input end **144** of second channel **114**. As hereinafter described, it can be appreciated that output end **128** of first channel **112** and input end **144** of second channel **114** may be utilized as valve **110** to terminate fluid flow from input spot **30** and output spot **32** along strip **40**.

In operation, reservoir **22** of microfluidic device **12** is filled with a selected fluid, such as oil **24**. One of more injectors **34** are configured to deliver input droplet **150** of a desired aqueous media on input spot **30**, outlet droplet **152** of a desired aqueous media on output spot **32**, and bridge droplet **154** so as to overlap output end **128** of first channel **112** and input end **144** of second channel **114**. It is intended for bridge droplet **154** to fluidically connect first and second channels **112** and **114**, respectively, thereby closing valve **110** and allowing for fluid flow therebetween.

In order to generate flow between input spot **30** and output spot **32** along strip **40**, it is contemplated to provide an external perturbation on the interface of oil **24** and strip **40** utilizing anti-static gun **50**. As heretofore described, oil **24** vibrates in response to the repetitive pumping of the ionized air by anti-static gun **50**, thus causing a momentum to be applied to the interface of oil **24** and strip **40**. This momentum helps the aqueous media overcome the energy barrier for the displacement of oil **24** and allows for the aqueous media to flow along strip **40** to fluidically connect input droplet **150** on input spot **30** to bridge droplet **154** and to fluidically connect bridge droplet **154** and output droplet **152** on output spot **32**. Because input droplet **150** has a smaller radius of curvature than bridge droplet **154**, a larger pressure exists on input spot **30**. The resulting pressure gradient causes aqueous media to flow from input droplet **150**, along first channel **112**, towards bridge droplet **154**. Similarly, bridge droplet **154** has a smaller radius of curvature than output droplet **152**, a larger pressure exists on input end **144** of second channel **114**. The resulting pressure gradient causes aqueous media to flow from bridge droplet **104**, along second channel **114**, towards output droplet **152**. It can be understood that by sequentially depositing additional drops **155** of aqueous media to input droplet **150** on input spot **30** with injector **34**, the resulting pressure gradient will cause the aqueous media to flow along first channel **112**, through bridge droplet **154**, along second channel **114** towards output droplet **152** on output spot **32**. As a result, fluid flows along strip **40** from input droplet **150** to output droplet **152**, FIG. **10**.

As heretofore described with respect to valve **80**, by terminating the depositing of drops **155** of aqueous media, it can be understood that the volume of aqueous media in bridge droplet **154** decreases thereby reducing the dimension thereof, FIGS. **11-12**. As the dimension of bridge droplet **154** is reduced to $V_{critical}$, output end **128** of first channel **112** becomes isolated from input end **144** of second channel **114**, thereby opening valve **110** and terminating the fluid flow between input spot **30** and output spot **32** along strip **40**. Reinstitution of fluid flow between first and second channels **112** and **114**, respectively, along strip **40** may be achieved by simply adding aqueous media to bridge droplet **154** to reestablish a fluidic connection between first and second channels **112** and **114**, respectively, and sequentially depositing additional drops **155** of aqueous media on input droplet **150**. Hence, by simply adding aqueous media to or removing aqueous media from bridge droplet **154**, connection or disconnection of valve **110** can be achieved reversibly.

Once again, it is noted that $V_{critical}$ is dependent upon the geometry and the size of the portion of ELR surface **28** between output end **128** of first channel **112** and input end **144** of second channel **114** (hereinafter referred to as the ELR gap). A larger ELR gap requires a larger $V_{critical}$ to maintain the connection between first and second channels **112** and **114**, respectively. The connection time ($\Delta t_{connection}$) of valve **1110** is defined as the time to reduce the initial volume of bridge droplet **154** ($V_{initial}$) to $V_{critical}$. It can be appreciated that a larger $V_{critical}$ (e.g., a valve with a larger ELR gap) results in a shortened for a given Q.

It is noted that fluid flow through valve **110** may be reversed by providing outlet droplet **152** with a smaller radius of curvature than bridge droplet **154** and by providing bridge droplet **1104** with a smaller radius of curvature than input droplet **150**. The resulting pressure gradient causes aqueous media to flow from output droplet **154**, along second channel **114**, towards bridge droplet **154**. Similarly, because bridge droplet **154** has a smaller radius of curvature than input droplet **150**, the resulting pressure gradient causes aqueous media to flow from bridge droplet **154**, along first channel **112**, towards input droplet **150**. It can be understood that by sequentially depositing additional drops **155** of aqueous media to outlet droplet **152**, the resulting pressure gradient will cause the aqueous media to flow along second channel **114**, through bridge droplet **154**, along first channel **112** towards input droplet **150**. As a result, fluid flows along strip **40** from output droplet **152** to input droplet **150**.

Referring to FIGS. **14-15**, it is contemplated for system **10** to include surface patterning provided on ELR surface **28** to allow for the generation of a gradient of particles between a first source region **230** to a second sink region **232** over a predetermined time period (the "gradient development period"). More specifically, in the depicted embodiment, hydrophilic source region **230** is pre-patterned on ELR surface **28** of microfluidic device **12** at a selected first location and a hydrophilic sink region **232** is pre-patterned on ELR surface **28** of microfluidic device **12** at a selected second location, axially spaced from the first location. Source region **230** has an outer periphery intersecting ELR surface **28** at boundary **235**. Similarly, sink region **232** has an outer periphery intersecting ELR surface **28** at boundary **238**. It is contemplated for sink region **232** to have a cross-sectional area generally equal to a cross-sectional area of source region **230**. However, source region **230** and sink region **232** may have other configurations without deviating from the scope of the present invention.

Source region **230** and sink region **232** are interconnected by a hydrophilic strip **240** pre-patterned on and extending axially along ELR surface **28**. Strip **240** is defined by first and second, generally parallel edges **242** and **244**, respectively, which intersect ELR surface **28**. First and second edges **242** and **244**, respectively, have corresponding first ends **242***a* and **244***a*, respectively, which intersect boundary **235** of source region **230** and corresponding second ends **242***b* and **244***b*, respectively, which intersect boundary **238** of sink region **32**. It is further contemplated for the surface patterning on ELR surface **28** to have other configurations. For example, strip **240** may be wider at second ends **242***b* and **244***b* and narrower at first ends **242***a* and **244***a* to promote passive pumping of an aqueous media in system **10**, as heretofore described. In addition, it is contemplated to provide source region **230** and sink region **232**, respectively, at different locations along strip **240**, without deviating from the scope of the present invention.

In operation, reservoir **22** of microfluidic device **12** is filled with a selected fluid, such as oil **24**. After flooding the reservoir **22** with oil **24**, one of more injectors **34**, FIG. **1**, are configured to deliver first input droplet (no shown) of a desired aqueous media on source region **230** and outlet

droplet **248** of a desired aqueous media on sink region **232**. Aqueous media is flowed along strip **240**, as heretofore described, to interconnect source region **230** and sink region **232**, thereby defining microchannel **241** along strip **240** having a height h, a width w and a length.

In order to generate a gradient of particles across microchannel **241** between source region **230** and sink region **232**, the convective flow of fluid in microchannel **241** must be minimized. It can be appreciated that a strong convective flow through microchannel **241** will transport the particles through, which reduces the ability to establish a gradient in microchannel **241** over an extended period of time. As such, to generate a gradient, it is necessary to minimize the pressure differential between the inlet of microchannel **241**, microchannel **241**, and the outlet of microchannel **241**. Fox example, to minimize the pressure differential between the inlet of microchannel **241**, microchannel **241**, and the outlet of microchannel **241**, the radii of the curvature of the droplets at the inlet of microchannel **24** and the outlet of microchannel must be maintained as close as possible. The combination of the size of source region **230** and sink region **232** patterned on ELR surface **28** and the volume of the droplets provided on source region **230** and sink region **232**, as hereinafter described, determine the local radius of curvature of the droplets. Consequently, it can be understood that size of source region **230** and the size of sink region **232** do not have to be the same, if the volumes of the droplet provided on source region **230** and sink region **232** are different.

In the depicted embodiment, one of more injectors **34** are configured to deliver second input droplet **252** of a desired aqueous media having a known concentration of particles, such as cells, molecules, chemical species, organisms or the like, on source region **230**. The particles in second input droplet **252** on source region **230** diffuse into microchannel **241** such that after a predetermined time period, a concentration gradient is created along the length of microchannel **241**. It can be understood that an ideal source/sink setup may be achieved by providing source and sink regions **230** and **232**, respectively, with volumes that are significantly larger that the volume of microchannel **241**. The large volume sink region **232** at the output end of microchannel **241** can help maintain the concentration gradient by not allowing the particles to accumulate in microchannel **241**. As such, the source/sink concept heretofore described may be used to create a pseudo-steady state in microchannel **241** wherein the concentration at a point within microchannel **241** does not vary dramatically with time.

Alternatively, it can be appreciated an ideal source/sink setup may be constructed by maintaining by a constant concentration of particles in microchannel **241** by providing an infinite source of particles at source region **230** and by providing a sink region **232** of infinite size. This may be accomplished by providing a steady flow through microchannel **241** utilizing a syringe pump to add volume to second inlet droplet **46**, while simultaneously removing the same volume of outlet droplet **248**.

Various modes of carrying out the invention are contemplated as being within the scope of the following claims particularly pointing out and distinctly claiming the subject matter that is regarded as the invention.

We claim:

1. An open microfluidic system, comprising:

a microfluidic device including a reservoir adapted for receiving an oil therein, the reservoir defined by a surface configured to repel an aqueous solution;

a hydrophilic input for receiving an input droplet of the aqueous solution thereon and a hydrophilic output for receiving an output droplet of the aqueous solution thereon, the input and the output being patterned on the surface and the output being spaced from the input;

a hydrophilic strip interconnecting the input and the output; and,

an injector configured to direct a series of input drops toward the input droplet so as to cause the flow of aqueous solution from the input droplet toward the output droplet along the hydrophilic strip.

2. The system of claim **1** wherein the hydrophilic strip includes a first channel having a first end connected to the input, a second channel having a first end connected to the output, and a valve configured to selectively fluidically connect the second ends of the first and second channel.

3. The system of claim **2** wherein the valve includes a second end of the first channel and a second end of the second channel.

4. The system of claim **3** wherein the valve includes a dried reagent fluidically interconnecting the second end of the first channel and the second end of the second channel, wherein fluid flowing over the dried reagent picks-up and re-dissolves the dried reagent therein, thereby exposing a portion of the surface between the first and second hydrophilic channels and fluidically isolating the first channel from the second channel.

5. The system of claim **3** wherein the second end of the second channel has a horseshoe configuration.

6. The system of claim **5** wherein the second end of the first channel has a horseshoe configuration.

7. An open microfluidic system, comprising a microfluidic device including a reservoir adapted for receiving an oil therein, the defined by a surface configured to repel an aqueous solution, a hydrophilic input and a hydrophilic output patterned on the surface, the output spaced from the input;

a hydrophilic strip interconnecting the input and the output, the hydrophilic strip including a first channel having a first end connected to the input, a second channel having a first end connected to the output, and a valve including a second end of the first channel and a second end of the second channel, the valve configured to selectively fluidically connect the second ends of the first and second channel;

and a droplet having a first dimension wherein the droplet communicates with the second end of the first channel and the second end of the second channel thereby closing the valve and a second dimension wherein the droplet fluidically isolates the second end of the first channel from the second end of the second channel thereby opening the valve.

8. The system of claim **1** wherein the hydrophilic strip includes an area having a reduced dimension.

9. A method of fabricating an open microfluidic system, comprising the steps: providing a microfluidic device including a reservoir defined by a surface configured to repel an aqueous solution; patterning a hydrophilic input, a hydrophilic output and a hydrophilic strip interconnecting the input and output on the surface; filling the reservoir with an oil; positioning an input droplet of the aqueous solution on the input and an output droplet of the aqueous solution on the output; fluidically connecting the input droplet and the output droplet along the hydrophilic strip with the aqueous solution; and directing a series of input drops toward the input droplet so as to cause the flow of the aqueous solution from the input droplet toward the outlet droplet along the hydrophilic strip.

10. A method of fabricating an open microfluidic system, comprising the steps:

providing a microfluidic device including a reservoir defined by a surface configured to repel an aqueous solution;

patterning a hydrophilic input, a hydrophilic output and a hydrophilic strip interconnecting the input and output on the surface;

filling the reservoir with an oil;

positioning an input droplet of the aqueous solution on the input and an output droplet of the aqueous solution on the output; and

fluidically connecting the input droplet and the output droplet along the strip with the aqueous solution;

wherein the step of fluidically connecting the input droplet and the output droplet along the strip includes the step of generating an external perturbation on the oil in the reservoir.

11. The method of claim 10 wherein the external perturbation on the oil is generated by an anti-static gun, the anti-static gun repetitively pumping ionized air at the oil.

12. The method of claim 9 wherein the hydrophilic strip includes a valve, the valve having a first open configuration fluidically isolating the input from the output and a second closed configuration wherein the input and the output are in fluidic communication.

13. The method of claim 12 wherein the hydrophilic strip includes a first channel has a first end connected to the input and a second channel having a first end connected to the output, the valve configured to selectively fluidically connect the first and second channels.

14. The method of claim 13 wherein the valve includes a second end of the first channel and a second end of the second channel.

15. The method of claim 14 wherein the valve includes a dried reagent fluidically interconnecting the second end of the first channel and the second end of the second channel.

16. The method of claim 15 comprising the additional step of a flowing a fluid over the dried reagent to picks-up and re-dissolve the dried reagent therein so as to expose a portion of the surface between the first and second hydrophilic channels and open the valve.

17. The method of claim 14 wherein the second end of the second channel has a horseshoe configuration.

18. The method of claim 17 wherein the second end of the first channel has a horseshoe configuration.

19. A method of fabricating an open microfluidic system, comprising the steps:

providing a reservoir defined by a surface configured to repel an aqueous solution;

patterning a hydrophilic input, a hydrophilic output and a hydrophilic strip interconnecting the input and output on the surface; filling the reservoir with an oil; positioning an input droplet of the aqueous solution on the input and an output the aqueous solution on the output; and fluidically connecting the input droplet and the output droplet along the hydrophilic strip with the aqueous solution, wherein:

the hydrophilic strip includes a valve, the valve having a first open configuration fluidically the input from the output and a second closed configuration wherein the input output are in fluidic communication; the hydrophilic strip includes a first channel having a first end connected to the input and a second having a first end connected to the output, the valve configured to selectively connect the first and second channels; the valve includes a second end of the first channel and a second end of the second the valve includes a droplet having a first dimension wherein the droplet communicates with the second end of the first channel and the second end of the second channel thereby closing the valve and a second dimension wherein the droplet fluidically isolates the second end of the first channel form the second end of the second channel thereby opening the valve.

20. The method of claim 9 including the steps of:

reducing a dimension of the hydrophilic strip so as to define a capture area; and

capturing a desired particle in the capture area.

21. The method of claim 9 including the of adjusting a dimension of the hydrophilic strip, wherein the dimension of the hydrophilic strip corresponds to a flow rate of the aqueous solution along the strip.

22. The method of claim 9 wherein the input droplet of the aqueous solution has a concentration of particles therein, the particles diffusing into the aqueous solution along the hydrophilic strip to form a gradient of particles in the aqueous solution along the hydrophilic strip.

23. A single-use valve, comprising:

a microfluidic device including a reservoir defined by a surface configured to repel an aqueous solution, the reservoir configured for receiving oil therein;

first and second hydrophilic channels patterned on surface, the first hydrophilic channel being spaced from and fluidically isolated from the second hydrophilic channel; and

a dried reagent deposited on the surface and interconnecting the first and second hydrophilic channels so as to fluidically connect the first and second hydrophilic channels;

wherein fluid flowing over the dried reagent picks-up and re-dissolves the dried reagent therein, thereby exposing a portion of the surface between the first and second hydrophilic channels, disconnecting the first and second hydrophilic channels, and fluidically isolating the first hydrophilic channel from the second hydrophilic channel.

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