[0006] In an aspect, the present disclosure provides a microfluidic device for processing a biological sample, comprising: a fluid flow path comprising a channel and an inlet port, ~~wherein the fluid flow path does not include an outlet port and~~ wherein the inlet port is configured to direct a solution comprising the biological sample to the channel, and wherein the channel defines at least a portion of the fluid flow path within which fluid flow is not hindered; and a chamber in fluid communication with the channel, wherein the chamber is configured to receive at least a portion of the solution from the channel and retain the at least the portion of the solution during the processing.

[0007] In some embodiments, the microfluidic device further comprises a plurality of chambers in fluid communication with the channel, wherein the plurality of chambers comprises the chamber. In some embodiments, the channel comprises a first end and a second end, and wherein the first end and the second end are connected to a single inlet port. In some embodiments, the fluid flow path is circular. In some embodiments, the channel comprises a first end and a second end, and wherein the first end is connected to the inlet port and the second end is connected to a ~~different inlet~~outlet port.

[0008] In some embodiments, the chamber is configured to permit pressurized off-gassing. In some embodiments, the chamber comprises a film or membrane that permits the pressurized off-gassing. In some embodiments, the film or membrane is polymer film or membrane. In some embodiments, the polymer film or membrane does not comprise an elastomer. In some embodiments, the film or membrane has a thickness of less than about 100 micrometers (μm). In some embodiments, the thickness is less than about 50 μm. In some embodiments, the film or membrane is substantially impermeable to liquids.

[0009] In some embodiments, the fluid flow path or the chamber does not include a valve. In some embodiments, a volume of the chamber is less than or equal to about 250 picoliters. In some embodiments, a volume of the chamber is less than or equal to about 500 picoliters. In some embodiments, the chamber has a cross-sectional dimension of less than or equal to about 250 μm. In some embodiments, the chamber has a depth of less than or equal to about 250 μm. In some embodiments, the microfluidic device further comprises a siphon aperture disposed between the channel and the chamber, wherein the siphon aperture is configured to provide fluid communication between the channel and the chamber.

[0010] In another aspect, the present disclosure provides methods for processing a biological sample, comprising: providing a device comprising (i) a fluid flow path comprising a channel

and an inlet port, ~~wherein the fluid flow path does not include an outlet port,~~ and (ii) a chamber in fluid communication with the channel; directing a solution comprising the biological sample from the inlet port to the channel, and wherein the channel defines at least a portion of the fluid flow path within which fluid flow is not hindered; and directing at least a portion of the solution from the channel to the chamber, which chamber retains the at least the portion of the solution during the processing.

[0011] In some embodiments, the device comprises a plurality of chambers in fluid communication with the channel, and wherein the plurality of chambers comprises the chamber. In some embodiments, the method further comprises applying a single pressure differential to the inlet port to direct solution from the inlet port to the channel and from the channel to the chamber. In some embodiments, the single pressure differential permits pressurized off-gassing of gas in the chamber.

[0012] In some embodiments, the method further comprises applying a first pressure differential to the inlet port to direct the solution from the inlet port to the channel. In some embodiments, the method further comprises applying a second pressure differential to the inlet port to direct the solution from the channel to the chamber. In some embodiments, the second pressure differential is greater than the first pressure differential. In some embodiments, the second pressure differential permits pressurized off-gassing of gas in the chamber. In some embodiments, the chamber comprises a film or membrane, and wherein the film or membrane permits pressurized off-gassing of the gas in the chamber.

[0013] In some embodiments, a volume of the solution is less than or equal to a volume of the chamber. In some embodiments, the device partitions the solution comprising the biological sample into the chamber such that no residual solution remains in the channel. In some embodiments, the method further comprises providing an immiscible fluid to the inlet port and directing the immiscible fluid to the channel. In some embodiments, a volume of the immiscible fluid is greater than a volume of the channel. In some embodiments, the biological sample is a nucleic acid molecule. In some embodiments, the method further comprises amplifying the nucleic acid molecule by thermal cycling the chamber. In some embodiments, the method further comprises controlling a temperature of the channel or the chamber. In some embodiments, the method further comprises detecting one or more components of the biological sample or a reaction with the one or more components of the biological sample in the chamber. In some embodiments, detecting the one or more components of the biological sample or the reaction comprises imaging the chamber.

[0014] ~~In another aspect, the present disclosure provides~~Disclosed herein are systems for processing a biological sample, comprising: a device comprising (i) a fluid flow path comprising a channel and an inlet port, wherein the fluid flow path does not include an outlet port, and wherein the inlet port is configured to direct a solution comprising the biological sample to the channel, and (ii) a chamber in fluid communication with the channel, wherein the chamber is configured to receive at least a portion of the solution from the channel and retain the at least the portion of the solution during the processing; a holder configured to receive and retain the device during the processing; and a fluid flow module configured to fluidically couple to the inlet port and supply a pressure differential to subject (i) the solution to flow from the inlet port to the channel and (ii) at least a portion of the solution to flow from the channel to the chamber.

[0015] In some embodiments, the device comprises a plurality of chambers in fluid communication with the channel, and wherein the plurality of chambers comprises the chamber. In some embodiments, the chamber of the device is configured to permit pressurized off-gassing of a gas in the chamber when the fluid flow module applies the pressure differential to the inlet port. In some embodiments, the chamber comprises a film or membrane that is configured to permit the pressurized off-gassing.

[0016] In some embodiments, the system further comprises one or more computer processors operatively coupled to the fluid flow module, wherein the one or more computer processors are individually or collectively programmed to direct the fluid flow module to supply the pressure differential when the fluid flow module is fluidically coupled to the inlet port, to thereby subject the solution to flow from the inlet port to the channel and direct the at least the portion of the solution from the channel to the chamber. In some embodiments, the system further comprises a thermal module in thermal communication with the chamber, wherein the thermal module is configured to control a temperature of the chamber during the processing. In some embodiments, the system further comprises a detection module in communication with the chamber, wherein the detection module is configured to detect a content of the chamber during the processing. In some embodiments, the detection module is an optical module in optical communication with the chamber. In some embodiments, the optical module is configured to image the chamber.

[0017] ~~In another aspect, the present disclosure provides~~Disclosed herein are systems for processing a biological sample, comprising a holder configured to retain a device comprising (i) a fluid flow path comprising a channel and an inlet port, wherein said fluid flow path does not include an outlet port, and (ii) a chamber in fluid communication with said channel; and one or more computer processors configured to be operatively coupled to said device when said device

is retained by said holder, wherein said one or more computer processors are individually or collectively programmed to (i) direct a solution comprising said biological sample from said inlet port to said channel; and (ii) direct at least a portion of said solution from said channel to said chamber, which chamber retains said at least said portion of said solution during said processing.

[0018] In some embodiments, the system further comprises a fluid flow module operatively coupled to said one or more computer processors, wherein said fluid flow module is configured to to be operatively coupled to said device when said device is retained by said holder, and wherein said one or more computer processors are programmed to direct said fluid flow module to direct said solution from said inlet port to said channel.

[0019] In some embodiments, the system further comprises a thermal module configured to be in thermal communication with said chamber when said device is retained by said holder, wherein said thermal module is configured to control a temperature of said chamber during said processing.

[0020] In some embodiments, the system further comprises a detection module configured to be in communication with said chamber when said device is retained by said holder, wherein said detection module is configured to detect a content of said chamber during said processing. In some embodiments, the detection module is an optical module in optical communication. In some embodiments, the optical module is configured to image said chamber.

[0021] Additional aspects and advantages of the present disclosure will become readily apparent to those skilled in this art from the following detailed description, wherein only illustrative embodiments of the present disclosure are shown and described. As will be realized, the present disclosure is capable of other and different embodiments, and its several details are capable of modifications in various obvious respects, all without departing from the disclosure. Accordingly, the drawings and description are to be regarded as illustrative in nature, and not as restrictive.

INCORPORATION BY REFERENCE

[0022] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference. To the extent publications and patents or patent applications incorporated by reference contradict the disclosure contained in the specification, the specification is intended to supersede and/or take precedence over any such contradictory material.

~~WHAT IS CLAIMED IS~~ **CLAIMS**:

1. A microfluidic device for processing a biological sample, comprising:

a fluid flow path comprising a channel and an inlet port, ~~wherein said fluid flow path does not include an outlet port, and~~ wherein said inlet port is configured to direct a solution comprising said biological sample to said channel, and wherein said channel defines at least a portion of the fluid flow path within which fluid flow is not hindered; and

a chamber in fluid communication with said channel, wherein said chamber is configured to receive at least a portion of said solution from said channel and retain said at least said portion of said solution during said processing.

2. The microfluidic device of claim 1, further comprising a plurality of chambers in fluid communication with said channel, wherein said plurality of chambers comprises said chamber.

3. The microfluidic device of claim 1, wherein said channel comprises a first end and a second end, and wherein said first end and said second end are connected to a single inlet port.

4. The microfluidic device of claim 3, wherein said fluid flow path is circular.

5. The microfluidic device of claim 1, wherein said channel comprises a first end and a second end, and wherein said first end is connected to said inlet port and said second end is connected to a ~~different inlet~~ outlet port.

6. The microfluidic device of claim 1, wherein said chamber is configured to permit pressurized off-gassing.

7. The microfluidic device of claim 6, wherein said chamber comprises a film or membrane that permits said pressurized off-gassing.

8. The microfluidic device of claim 7, wherein said film or membrane is a polymer film or membrane.

9. The microfluidic device of claim 8, wherein said polymer film or membrane does not comprise an elastomer.

10. The microfluidic device of claim 7, wherein said film or membrane has a thickness of less than about 100 micrometers (μm).

11. The microfluidic device of claim 10, wherein said thickness is less than about 50 μm.

12. The microfluidic device of claim 7, wherein said film or membrane is substantially impermeable to liquids.

13. The microfluidic device of claim 1, wherein said fluid flow path or said chamber does not include a valve.

14. The microfluidic device of claim 1, wherein a volume of said chamber is less than or equal to about 500 picoliters.

15. The microfluidic device of claim 1, wherein a volume of said chamber is less than or equal to about 250 picoliters.

16. The microfluidic device of claim 1, wherein said chamber has a cross-sectional dimension of less than or equal to about 250 μm.

17. The microfluidic device of claim 1, wherein said chamber has a depth of less than or equal to about 250 μm.

18. The microfluidic device of claim 1, further comprising a siphon aperture disposed between said channel and said chamber, wherein said siphon aperture is configured to provide fluid communication between said channel and said chamber.

19. The microfluid device of claim 1, further comprising a sacrificial chamber that is in fluid communication with said channel.

20. The microfluid device of claim 19, wherein said sacrificial chamber is configured to retain an excess portion of a solution comprising said biological sample.

21. A method for processing a biological sample, comprising:

(a) providing a device comprising (i) a fluid flow path comprising a channel and an inlet port, ~~wherein said fluid flow path does not include an outlet port,~~ and (ii) a chamber in fluid communication with said channel;

(b) directing a solution comprising said biological sample from said inlet port to said channel, <u>wherein said channel defines at least a portion of the fluid flow path within which fluid flow is not hindered</u>; and

(c) directing at least a portion of said solution from said channel to said chamber, which chamber retains said at least said portion of said solution during said processing.

22. The method of claim 21, wherein said device comprises a plurality of chambers in fluid communication with said channel, and wherein said plurality of chambers comprises said chamber.

23. The method of claim 21, further comprising applying a single pressure differential to said inlet port to direct said solution from said inlet port to said channel and from said channel to said chamber.

24. The method of claim 23, wherein said single pressure differential permits pressurized off-gassing of gas in said chamber.

25. The method of claim 21, further comprising applying a first pressure differential to said inlet port to direct said solution from said inlet port to said channel.

26. The method of claim 25, further comprising applying a second pressure differential to said inlet port to direct said solution from said channel to said chamber.

27. The method of claim 26, wherein said second pressure differential is greater than said first pressure differential.

28. The method of claim 26, wherein said second pressure differential permits pressurized off-gassing of gas in said chamber.

29. The method of claim 24 or claim 28, wherein said chamber comprises a film or membrane, and wherein said film or membrane permits pressurized off-gassing of said gas in said chamber.

30. The method of claim 21, wherein a volume of said solution is less than or equal to a volume of said chamber.

31. The method of claim 30, wherein said device partitions said solution comprising said biological sample into said chamber such that no residual solution remains in said channel.

32. The method of claim 21, further comprising providing an immiscible fluid to said inlet port and directing said immiscible fluid to said channel.

33. The method of claim 32, wherein a volume of said immiscible fluid is greater than a volume of said channel.

34. The method of claim 21, wherein said biological sample is a nucleic acid molecule.

35. The method of claim 34, further comprising amplifying said nucleic acid molecule by thermal cycling said chamber.

36. The method of claim 21, further comprising controlling a temperature of said channel or said chamber.

37. The method of claim 21, further comprising detecting one or more components of said biological sample or a reaction with said one or more components of said biological sample in said chamber.

38. The method of claim 37, wherein detecting said one or more components of said biological sample or said reaction comprises imaging said chamber.

~~39. A system for processing a biological sample, comprising:~~

~~a device comprising (i) a fluid flow path comprising a channel and an inlet port, wherein said fluid flow path does not include an outlet port, and wherein said inlet port is configured to direct a solution comprising said biological sample to said channel, and (ii) a chamber in fluid communication with said channel, wherein said chamber is configured to receive at least a portion of said solution from said channel and retain said at least said portion of said solution during said processing;~~

~~a holder configured to receive or retain said device during said processing; and~~

~~a fluid flow module configured to fluidically couple to said inlet port and supply a pressure differential to subject (i) said solution to flow from said inlet port to said channel and (ii) at least a portion of said solution to flow from said channel to said chamber.~~

~~40. The system of claim 39, wherein said device comprises a plurality of chambers in fluid communication with said channel, and wherein said plurality of chambers comprises said chamber.~~

~~41. The system of claim 39, wherein said chamber of said device is configured to permit pressurized off-gassing of a gas in said chamber when said fluid flow module applies said pressure differential to said inlet port.~~

~~42. The system of claim 41, wherein said chamber comprises a film or membrane that is configured to permit said pressurized off-gassing.~~

~~43. The system of claim 39, further comprising one or more computer processors operatively coupled to said fluid flow module, wherein said one or more computer processors are individually or collectively programmed to direct said fluid flow~~

module to supply said pressure differential when said fluid flow module is fluidically coupled to said inlet port, to thereby subject said solution to flow from said inlet port to said channel and direct said at least said portion of said solution from said channel to said chamber.

44. The system of claim 39, further comprising a thermal module in thermal communication with said chamber, wherein said thermal module is configured to control a temperature of said chamber during said processing.

45. The system of claim 39, further comprising a detection module in communication with said chamber, wherein said detection module is configured to detect a content of said chamber during said processing.

46. The system of claim 45, wherein said detection module is an optical module in optical communication with said chamber.

47. The system of claim 48, wherein said optical module is configured to image said chamber.

48. A system for processing a biological sample, comprising:

a holder configured to retain a device comprising (i) a fluid flow path comprising a channel and an inlet port, wherein said fluid flow path does not include an outlet port, and (ii) a chamber in fluid communication with said channel; and

one or more computer processors configured to be operatively coupled to said device when said device is retained by said holder, wherein said one or more computer processors are individually or collectively programmed to (i) direct a solution comprising said biological sample from said inlet port to said channel; and (ii) direct at least a portion of said solution from said channel to said chamber, which chamber retains said at least said portion of said solution during said processing.

49. The system of claim 48, further comprising a fluid flow module operatively coupled to said one or more computer processors, wherein said fluid flow module is configured to to be operatively coupled to said device when said device is retained by said holder, and wherein said one or more computer processors are programmed to direct said fluid flow module to direct said solution from said inlet port to said channel.

50. The system of claim 48, further comprising a thermal module configured to be in thermal communication with said chamber when said device is retained by said

~~holder, wherein said thermal module is configured to control a temperature of said chamber during said processing.~~

~~51. The system of claim 48, further comprising a detection module configured to be in communication with said chamber when said device is retained by said holder, wherein said detection module is configured to detect a content of said chamber during said processing.~~

~~52. The system of claim 51, wherein said detection module is an optical module in optical communication.~~

~~53. The system of claim 52, wherein said optical module is configured to image said chamber.~~