

CLAIMS

We claim:

1. ~~1. An array assembly, comprising:
a first layer and at least a second layer;
a third layer having a plurality of wells formed therein, the third layer being disposed between the first and second layers;
a vacuum channel in fluid communication with the plurality of wells and with a vacuum port;
and
an array fill channel in fluid communication with the plurality of wells and with a fluid source.~~
2. ~~2. The array assembly of claim 1, wherein the first and second layers are film layers.~~
3. ~~3. The array assembly of claim 1, wherein the third layer comprises a card layer having the plurality of wells formed therein.~~
4. ~~4. The array assembly of claim 3, wherein the first layer is bonded to a first side of the card layer and seals a first end of each of the plurality of wells, and the second layer is bonded to a second side of the card layer and seals a second end of each of the plurality of wells.~~
5. ~~5. The array assembly of claim 1, wherein the vacuum port comprises an opening in one or more of the first layer or the second layer.~~
6. ~~6. The array assembly of claim 3, further comprising a recessed conduit in the card layer, wherein at least a portion of one or more of the vacuum channel and the array fill channel is formed between the second film layer and the recessed conduit in the card layer, and wherein the recessed conduit in the card layer has a manifold configuration that fluidically connects the plurality of wells to the vacuum port by at least two paths and fluidically connects each well to the fluid source by at least two paths.~~
7. ~~7. The array assembly of claim 1, wherein at least a portion of the vacuum channel is disposed in and/or co-localized with at least a portion of the array fill channel.~~
8. ~~8. The array assembly of claim 1, further comprising an openable seal in the array fill channel between the plurality of wells and the fluid source.~~
9. ~~9. The array assembly of claim 1, wherein the plurality of wells are disposed in a respective plurality of rows, the array fill channel comprises an access channel in fluid communication with the fluid source and with a manifold channel assembly, the manifold channel assembly comprising:
a plurality of branch channels extending along the respective plurality of rows, the plurality of branch channels respectively being in fluid communication with the plurality of wells; and
a first main channel extending along a first end of the plurality of rows, the plurality of branch channels extending from the first main channel, the array fill channel in communication with the first main channel.~~
10. ~~10. The array assembly of claim 9, wherein the manifold channel assembly further comprises a second main channel in fluid communication with the plurality of branch channels, the second main channel extending along a second end of the plurality of rows, the vacuum port in~~

~~communication with the second main channel, wherein each well is in fluid communication with the vacuum port and the fluid source by at least two different paths.~~

~~11. — The array assembly of claim 10, wherein the manifold channel assembly further comprises a plurality of connection channels extending respectively from each of the plurality of branch channels to the plurality of wells, the plurality of connection channels respectively being in fluid communication with the plurality of wells.~~

~~12. — A method of manufacturing an array assembly, the method comprising:~~

~~providing a card layer having a plurality of wells disposed therein;~~

~~disposing the card layer between a first film layer and at least a second film layer;~~

~~bonding the first film layer to a first side of a card layer; and~~

~~bonding the second film layer to a second side of the card layer,~~

~~wherein the card layer and at least one of the first or second film layers form:~~

~~(i) a vacuum channel extending between and/or in fluid communication with the plurality of wells and a vacuum port; and~~

~~(ii) an array fill channel extending between and/or in fluid communication with the plurality of wells and a fluid source.~~

~~13. — The method of claim 12, wherein the recessed conduit in the card layer has a manifold configuration.~~

~~141. A method of using an array assembly on a sample, comprising~~

~~(a) providing a sample container comprising a reaction zone in fluid communication with the array assembly, the array assembly comprising~~

~~a plurality of wells configured in an array,~~

~~an access opening between the reaction zone and array,~~

~~a vacuum port, and~~

~~a plurality of channels such that each well in the array is fluidly connected to the access opening and the vacuum port;~~

~~(b) performing an analytical method on the sample in the reaction zone to produce a reaction mixture;~~

~~(c) opening the vacuum port and drawing a vacuum on the array assembly such that air is evacuated from the plurality of wells and the plurality of channels;~~

~~(d) sealing the vacuum port such that the plurality of wells are maintained under the vacuum, thereby forming an evacuated array; and~~

~~(e) opening the access opening such that the reaction mixture is drawn into the plurality of wells via the array fill plurality of channel.~~

~~152. The method of claim 14, further comprising mixing the reaction mixture with one or more reagents disposed in each of the plurality of wells to form a second mixture, and performing a second analytical method on the second reaction mixture.~~

~~163.~~ The method of claim ~~152~~, wherein the second mixture is a PCR mixture.

~~174.~~ The method of claim ~~163~~, further comprising thermocycling the second reaction mixture.

~~185.~~ The method of claim ~~174~~, further comprising detecting an amplification product in at least one of the plurality of wells.

~~196.~~ The method of claim ~~152~~, wherein the sample comprises a microorganism and the one or more reagents comprises an antibiotic.

~~207.~~ The method of claim ~~196~~, wherein the antibiotic is present in a first of the plurality of wells at a first concentration and is present in a second of the plurality of wells at a second concentration, wherein the second concentration is lower than the first concentration.

~~218.~~ The method of claim 14, wherein step (c) provides a vacuum in the plurality of wells of between about 2 millibar and about 150 millibar.

~~229.~~ The method of claim 14, wherein steps (c) and (d) are performed before step (b) is completed.

~~23.~~—A method for performing a multi-step biological reaction in a sealed sample container, comprising:

~~(a) providing a sealed container comprising a first reaction zone, a second reaction zone, and dried components in the second reaction zone,~~

~~(b) performing a first reaction in the first reaction zone, to generate a reaction mixture,~~

~~(c) hydrating the dried components in the second reaction zone with a hydration fluid to generate hydrated components, wherein step (c) is performed before or during step (b),~~

~~(d) adding a portion of the reaction mixture to the hydrated components, and~~

~~(e) performing a second reaction in the second reaction zone.~~

~~24.~~—The method of claim ~~23~~, wherein the second reaction zone is an array of wells, wherein each well is provided with dried components, and wherein at least a plurality of wells has components that are different from other wells in the plurality of wells.

~~25.~~—The method of claim ~~24~~, wherein each individual well in the array of wells is provided with a barrier layer that retards entry of the hydration fluid into the wells.

~~26.~~—The method of claim ~~25~~, wherein the hydrating step comprises filling the plurality of wells with the hydration fluid, followed by removing from the array of wells of any excess hydration fluid.

~~27.~~—The method of claim ~~26~~, wherein the adding step comprises

~~moving the reaction mixture to the array,~~

~~allowing a portion of the reaction mixture to cross the barrier layer and enter each well, and~~

~~removing from the array any excess reaction mixture.~~

~~28.~~—The method of claim ~~26~~, wherein the adding step comprises

~~moving the reaction mixture to the array, and~~

~~sealing a portion of the reaction mixture adjacent each well outside the barrier layer, so that the reaction mixture and hydrated components may mix through the barrier layer.~~

~~29. — The method of claim 25, wherein the barrier layer is a pierced layer having one or more openings per well, wherein the openings are sized such that absent some force, fluid does not readily flow through the openings.~~

~~30. — The method of claim 24, wherein the first reaction is first stage PCR and the second reaction is second PCR.~~

~~31. — The method of claim 30, wherein the second reaction zone is maintained at a cool temperature prior to step (d).~~

~~32. — The method of claim 23, further comprising evacuating at least the second reaction zone with a partial vacuum prior to step (c).~~

~~33. — An array assembly, comprising:~~

~~a card having a plurality of wells arranged in an array;~~

~~an access opening in a first side of the card;~~

~~a channel system in fluid communication with the access opening and with the plurality of wells; and~~

~~a vacuum port in fluid communication with the channel system.~~

~~34. — The array assembly of claim 33, wherein the array comprises a plurality of rows, each of the plurality of rows comprising one or more wells, the channel system comprising a manifold channel assembly comprising:~~

~~a plurality of branch channels extending along the plurality of rows, the plurality of branch channels respectively being in fluid communication with the plurality of wells; and~~

~~a first main channel in fluid communication with the access opening extending along a first end of the plurality of rows, the plurality of branch channels extending from the first main channel;~~

~~a second main channel in fluid communication with the plurality of branch channels, the second main channel extending along a second end of the plurality of rows, the vacuum port in communication with the second main channel;~~

~~wherein each well is in fluid communication with the vacuum port and the access opening by at least two different paths.~~

~~35. — The array assembly of one of claims 33 or 34, further comprising a first film layer and a second film layer, wherein first and second film layers form a pouch, the card layer being at least partially sealed inside the pouch.~~

~~36. — A reaction container, comprising:~~

~~a plurality of fluidly connected reaction chambers;~~

~~an array comprising~~

~~an access opening in fluid communication with at least one of the fluidly connected reaction chambers;~~

~~a plurality of reaction wells;~~

~~a vacuum port;~~

~~a channel system in fluid communication with the access opening, with the plurality of wells and with the vacuum port, the channel system providing paths such that each reaction well in the array is fluidly connected to the vacuum port by at least two paths and each reaction well is also connected to the fluid source by at least two paths.~~

~~37. — The array assembly of claim 36, wherein the vacuum port is reversibly sealable and the access opening is reversibly sealable.~~

~~38. — An array assembly, comprising:~~

~~a plurality of wells arranged in an array;~~

~~a vacuum channel in fluid communication with the plurality of wells and with a vacuum port;~~
and

~~an array fill channel in fluid communication with the plurality of wells and with a fluid source.~~

~~39. — The array assembly of claim 38, further comprising~~

~~a card layer having the plurality of wells formed therein,~~

~~a first film layer bonded to a first side of the card layer, wherein the first film layer seals a first end of each of the plurality of wells;~~

~~a second film layer bonded to a second side of the card layer, wherein the vacuum port comprises an opening in one or more of the first film layer and the second film layer.~~

~~40. — The array assembly of claim 39, wherein at least a portion of one or more of the vacuum channel and the array fill channel is formed (i) in the card layer, (ii) in the second film layer, or (iii) between the card layer and the second film layer.~~

~~41. — The array assembly of claim 39, wherein at least a portion of one or more of the vacuum channel and the array fill channel is formed between the first film layer and the second film layer.~~

~~42. — The array assembly of claim 39, wherein a portion of the array fill channel is formed between the first film layer and the second film layer and is disposed about at least a portion of a perimeter of the card layer.~~

~~43. — The array assembly of claim 39, further comprising a recessed conduit in the card layer, wherein the recessed conduit in the card layer has a manifold configuration wherein each well in the array is fluidly connected to the vacuum port by at least two paths and each well is also connected to the fluid source by at least two paths.~~

~~44. — The array assembly of claim 43, wherein the second film layer has a recessed conduit formed therein, further comprising a third film layer disposed between the second film layer and the card layer, wherein the third film layer has a plurality of piercings extending therethrough, and wherein the plurality of piercings are in fluid communication with the recessed conduit in the second film layer.~~

~~45. — The array assembly of claim 44, wherein at least a portion of the array fill channel is disposed between the recessed conduit in the second film layer and the bond between the third film~~

layer and the second film layer between two or more portions of the recessed conduit in the second film layer.

46. — The array assembly of claim 38, wherein at least a portion of the vacuum channel is provided as a portion of the array fill channel and is configured to maintain the array fill channel in an open position.

47. — The array assembly of claim 38, further comprising an openable seal in the array fill channel between the plurality of wells and the fluid source.

48. — The array assembly of claim 38, further comprising one or more reagents disposed in each of the plurality of wells.

49. — The array assembly of claim 38, wherein the plurality of wells are disposed in a respective plurality of rows, the array fill channel comprising an access channel in fluid communication with the fluid source and with a manifold channel assembly, the manifold channel assembly comprising:

a plurality of branch channels extending along the respective plurality of row, the plurality of branch channels respectively being in fluid communication with the plurality of wells; and

a first main channel extending along a first end of the plurality of rows, the plurality of branch channels extending from the first main channel, the array fill channel extending from the first main channel.

50. — The array assembly of claim 49, wherein the manifold channel assembly further comprises a second main channel in fluid communication with the plurality of branch channels, the second main channel extending along a second end of the plurality of rows, the vacuum port in communication with the second main channel, wherein each well is in fluid communication with the vacuum port and the fluid source by at least two different paths.

51. — The array assembly of claim 49, wherein the manifold channel assembly further comprises a plurality of connection channels extending respectively from each of the plurality of branch channels to the plurality of wells, the plurality of connection channels respectively being in fluid communication with the plurality of wells.

5210. A method of drawing a vacuum on a reaction container *in situ* while performing an analytical method, the method comprising:

providing a reaction container that includes a sample introduction zone, at least a first reaction zone in fluid communication with the sample introduction zone, and a second reaction zone in fluid communication with the first reaction zone, wherein the second reaction zone comprises

a plurality of wells,

a vacuum channel in fluid communication with the plurality of wells and a vacuum port,

an array fill channel extending between and/or in fluid communication with the plurality of wells and the first reaction zone, and

an openable seal disposed between the first reaction zone and the second reaction zone;

performing at least one step of the analytical method with the reaction container;

drawing a partial vacuum on the plurality of wells such that the plurality of wells, the vacuum channel, and at least a portion of the array fill channel are under reduced pressure relative to atmospheric pressure;

sealing a portion of the vacuum channel such that the plurality of wells and at least the portion of the array fill channel are maintained under the vacuum, thereby forming an evacuated array; and

applying a fluid to the evacuated array by opening the openable seal disposed between the first reaction zone and the second reaction zone such that the fluid is drawn into the plurality of wells via the array fill channel.

5311. The method of claim 5210, wherein the at least one step of the analytical method includes removing the reaction container from an ambient pressure package, introducing a sample into the sample introduction zone, inserting the reaction container into an instrument configured for performing the analytical method and configured for drawing a partial vacuum to form the evacuated array, performing one or more reactions in the first reaction zone, or preparing for applying the fluid to the evacuated array.

5412. The method of claim 5311, wherein performing one or more reactions in the first reaction zone comprises one or more of performing a sample lysis to generate a lysate, isolating lysis particles from the lysate, mixing silica magnetic beads with the lysate, moving a residual lysate after nucleic acid capture with the silica magnetic beads to a waste chamber, performing at least one wash of the magnetic beads and moving the wash liquid to the waste chamber, eluting nucleic acids from the silica magnetic beads, performing a first singleplex or multiplex PCR reaction, or diluting a product of the first PCR reaction in preparation for performing a second PCR reaction in the plurality of wells of the second reaction zone.

5513. The method of claim 5210, wherein the reaction container further includes one or more reagent blisters fluidically connected to one or more of the sample introduction zone, the first reaction zone, or the second reaction zone.

5614. The method of claim 5513, wherein at least one of the one or more reagent blisters includes a dried reagent, and the method further comprising drawing a partial vacuum on the one or more reagent blisters including the dried reagent.

5715. The method of claim 5614, further comprising mixing a fluid with the dried reagent to form a first mixture.

5816. The method of claim 5614, wherein the one or more reagent blisters comprise reagents for sample preparation, nucleic acid recovery, a first-stage PCR, and a second-stage PCR.

5917. A system, comprising:

a reaction container that includes a sample introduction zone, at least a first reaction zone in fluid communication with the sample introduction zone, and a second reaction zone in fluid communication with the first reaction zone, wherein the second reaction zone comprises

a plurality of wells,

a vacuum channel extending between and/or in fluid communication with the plurality of wells and a vacuum port,

an array fill channel extending between and/or in fluid communication with the plurality of wells and the first reaction zone, and

an openable seal disposed between the first reaction zone and the second reaction zone; and

an instrument configured to perform an analytical method using the reaction container, wherein the instrument includes a vacuum system to draw a partial vacuum in one or more portions of the reaction container while performing one or more steps of the analytical method.

6018. The system of claim 5917, wherein the one or more portions of the reaction container configured to have a partial vacuum drawn thereon are substantially dry such that the partial vacuum is not being drawn against a partial pressure of water.

6119. The system of claim 6018, wherein the partial vacuum in the one or more portions of the reaction container configured to have a partial vacuum drawn thereon is in a range between about 2 millibar and about 150 millibar.

6220. The system of claim 5917, wherein the one or more portions of the reaction container configured to have a partial vacuum drawn thereon include the plurality of wells, the vacuum channel, and at least a portion of the array fill channel.

6321. The system of claim 5917, wherein the instrument includes a seal device to apply a seal to preserve the partial vacuum drawn in the one or more portions of the reaction container.

6422. The system of claim 5917, wherein the reaction container further includes one or more reagent blisters fluidically connected to one or more of the sample introduction zone, the first reaction zone, or the second reaction zone.

6523. The system of claim 6422, wherein at least one of the one or more reagent blisters includes a dried reagent, and wherein the one or more portions of the reaction container configured to have a partial vacuum drawn thereon include the one or more reagent blisters that include the dried reagent.

6624. The system of claim 5917, wherein the instrument is a PCR instrument that includes at least one heater positioned and arranged for thermocycling at least one portion of the reaction container.

6725. The system of claim 5917, wherein the instrument includes at least one heater positioned and arranged for controlling the temperature in at least one portion of the reaction container for performing an isothermal reaction.

6826. The system of claim 5917, wherein the instrument includes at least one heater positioned and arranged for controlling the temperature in at least one portion of the reaction container, one or more actuators for moving fluid in the reaction container, and one or more seals for controlling the movement of fluids within one or more portions of the reaction container.