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(54) **Title**: MICROFLUIDIC CARTRIDGE FOR PROCESSING AND DETECTING NUCLEIC ACIDS

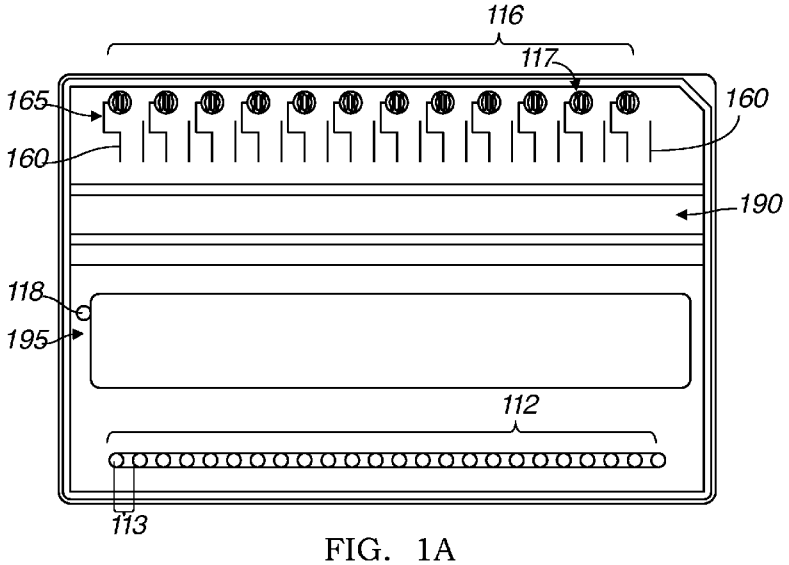

FIG. 1A

(57) **Abstract**: A microfluidic cartridge, configured to facilitate processing and detection of nucleic acids, comprising: a top layer comprising a set of cartridge-aligning indentations, a set of sample port-reagent port pairs, a shared fluid port, a vent region, a heating region, and a set of Detection chambers; an intermediate substrate, coupled to the top layer comprising a waste chamber; an elastomeric layer, partially situated on the intermediate substrate; and a set of fluidic pathways, each formed by at least a portion of the top layer and a portion of the elastomeric layer.

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1. A cartridge, configured to facilitate processing and detecting of a nucleic acid, comprising:
   - a first layer comprising a sample port , a reagent port, a fluid port, and a detection chamber;
   - an elastomeric layer; and
   - a fluidic pathway, formed by at least a portion of the first layer and a portion of the elastomeric layer, wherein the fluidic pathway is fluidically coupled to the sample port, the reagent port, the fluid port, and the detection chamber, and is configured to be occluded at a set of occlusion positions upon manipulation of the elastomeric layer,
     - wherein a first truncated pathway is defined upon manipulation of the fluidic pathway at a first subset of the set of occlusion positions, and
     - wherein a second truncated pathway to the detection chamber is defined upon manipulation of the fluidic pathway at a second subset of the set of occlusion positions.

2. The cartridge of Claim 1, wherein the first layer further comprises a vent region including a liquid-impermeable membrane, and wherein the fluidic pathway is configured to pass through the vent region upstream of the detection chamber.

3. The cartridge of Claim 1, further comprising an intermediate substrate, coupled to the first layer and defining a waste chamber and a set of valve guides, wherein the waste chamber has a corrugated surface, such that the corrugated surface defines a region configured to receive a magnet providing a magnetic field configured to trap nucleic acids bound to magnetic beads within the first truncated pathway.

4. The cartridge of Claim 1, wherein a third truncated pathway, coupled to the fluid port, not coupled to the sample port, and not coupled to the reagent port, is defined upon occlusion of the branched fluidic pathway at a third subset of the set occlusion positions, wherein the first subset of the set of occlusion positions, the second subset of the set of occlusion positions, and the third subset of the set of occlusion positions are overlapping, and wherein the set of occlusion positions comprises at least one normally open position and one normally closed position.

5. A cartridge, configured to facilitate processing of a nucleic acid, comprising:
   - a sample port, a reagent port, and a fluid port;
   - a magnet region configured to receive a magnet; and

- a fluidic pathway, wherein the fluidic pathway is fluidically coupled to the sample port, the reagent port, and the fluid port, and comprises a capture segment configured to cross the magnet region, wherein the capture segment is a turnabout segment configured to cross the magnet region and configured to trap nucleic acids bound to magnetic beads by way of a magnetic field provided by the magnet.

6. The cartridge of Claim 5, further comprising a first layer, an intermediate substrate, and an elastomeric layer,

- wherein the first layer defines the sample port, the reagent port, the fluid port, and a detection chamber,
- wherein the intermediate substrate is configured to couple to the first layer, such that the elastomeric layer is situated between the intermediate substrate and the first layer, and wherein the intermediate substrate defines the magnet region, and
- wherein the fluidic pathway is formed by at least a portion of the first layer and a portion of the elastomeric layer, and is configured to be occluded upon manipulation of the elastomeric layer.

7. The cartridge of Claim 6, wherein the intermediate substrate is partially separated from the first layer by a film layer, and wherein the intermediate substrate further defines a waste chamber configured to partially form the magnet region, a waste inlet coupled to the fluidic pathway, and a waste vent.

8. The cartridge of Claim 6, further comprising a vent region and a heating region, such that the capture segment of the fluidic pathway is configured to cross the magnet region and the heating region, and the fluidic pathway is configured to pass through the vent region upstream of the detection chamber.

9. A cartridge, configured to facilitate processing of nucleic acids, comprising:

- a shared fluid port configured to receive a processing fluid;
- a region accessible by a magnet providing a magnetic field;
- a first fluidic pathway for processing a first sample, wherein the first fluidic pathway is coupled to the shared fluid port and comprises a first capture segment configured to cross the region; and

- a second fluidic pathway for processing a second sample, wherein the second fluidic pathway is coupled to the shared fluid port and comprises a second capture segment configured to cross the magnet region.

10. The cartridge of Claim 9, further comprising a first layer, an intermediate substrate, and an elastomeric layer,

- wherein the first layer defines a first sample port-reagent port pair, a second sample port-reagent port pair, the shared fluid port, a first detection chamber, and a second detection chamber,
- wherein the first layer further comprises a vent region including a liquid-impermeable film, such that the first fluidic pathway is configured to pass through the vent region upstream of the first detection chamber, and the second fluidic pathway is configured to pass through the vent region upstream of the second detection chamber,
- wherein the elastomeric layer is situated between the intermediate substrate and the first layer,
- wherein the first fluidic pathway is formed by at least a portion of the first layer and a portion of the elastomeric layer, and is coupled to the first sample port-reagent port pair and to the first detection chamber, and
- wherein the second fluidic pathway is formed by at least a portion of the first layer and a portion of the elastomeric layer, and is coupled to the second sample port-reagent port pair and to the second detection chamber.

11. The cartridge of Claim 10, wherein the intermediate substrate further defines a waste chamber configured to receive a waste fluid from at least one of the first fluidic pathway and the second fluidic pathway, and wherein the waste chamber comprises a corrugated surface, such that the region accessible by the magnet is defined by the corrugated surface.

12. The cartridge of Claim 9, wherein the first capture segment is an s-shaped segment with an aspect ratio less than one, and the second capture segment is an s-shaped segment with an aspect ratio less than one.

13. A cartridge, configured to facilitate processing and detecting of nucleic acids, comprising:

- a first layer and an intermediate substrate, coupled to the first layer, wherein the intermediate substrate defines a waste chamber;

- a first fluidic pathway, formed by at least a portion of the first layer; and
- a second fluidic pathway, formed by at least a portion of the first layer, wherein the first fluidic pathway is configured to transfer waste to the waste chamber and wherein the second fluidic pathway is configured to transfer waste to the waste chamber.

14. (CURRENTLY AMENDED) The cartridge of Claim 13, wherein the first layer comprises a first sample port-reagent port pair, a second sample port-reagent port pair, a shared fluid port, a first detection chamber, and a second detection chamber, wherein the first fluidic pathway is coupled to the first sample port-reagent port pair and the first detection chamber, wherein the second fluidic pathway is coupled to the second sample port-reagent port pair and the second detection chamber, and wherein the shared fluid port is configured to fluidically couple to the first fluidic pathway and the second fluidic pathway.

15. The cartridge of any one of Claims 9 and 14, further comprising a heating region and a vent region, such that the first fluidic pathway is configured to cross the heating region and to pass through the vent region upstream of the first detection chamber, and the second fluidic pathway is configured to cross the heating region and to pass through the vent region upstream of the second detection chamber, and wherein the heating region is defined by a recessed region of the cartridge and spans a long dimension of the cartridge.

16. The cartridge of any one of Claims 8 and 15, wherein the vent region comprises a hydrophobic, liquid impermeable membrane that is gas permeable.

17. The cartridge of Claim 14, wherein the first detection chamber comprises a first serpentine-shaped fluidic channel, and the second detection chamber comprises a second serpentine-shaped fluidic channel, and wherein at least one of the first serpentine-shaped fluidic channel and the second serpentine-shaped fluidic channel comprises three wide channels interconnected by two narrow channels.

18. The cartridge of Claim 14, wherein the first detection chamber and the second detection chamber are identical, wherein the first detection chamber is configured to be heated from one side and the second detection chamber is configured to be heated from one side, and wherein at least one of the first detection chamber and the second detection chamber is configured to be optimized for volumetric capacity, thermocycling rates, optical detection, and filling in a manner that limits

bubble generation.

19. The cartridge of Claim 13, wherein the waste chamber comprises a corrugated surface defining a magnet housing region, and wherein the waste chamber is located below the first layer.

20. The cartridge of any one of Claims 1, 5, 9, and 13, wherein the cartridge adheres to microtiter plate dimensional standards.