

INTEGRATED MULTIPLEX TARGET ANALYSIS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of provisional patent application Ser. No. 61/717,887, filed Oct. 24, 2012, and Ser. No. 61/798,091, filed Mar. 15, 2013, both of which are hereby incorporated by reference in their entirety, and particularly for the Figures and Legends contained therein.

BACKGROUND OF THE INVENTION

[0002] One major challenge in the area of clinical and molecular diagnostics is the ability to have a “sample to answer” system that allows minimal sample handling and preparation, rapid assays as well as no requirement for highly trained laboratory personnel. While many systems have been proposed, to date there are virtually no such commercial systems. The present invention provides such an integrated, multiplex system.

BRIEF SUMMARY OF THE INVENTION

[0003] The present invention provides biochip cartridges and instrument devices for the detection and/or analysis of target analytes from patient samples.

[0004] Accordingly, in one aspect, the present invention provides biochip cartridges generally comprising a bottom substrate and a top plate. The bottom substrate comprises a printed circuit board (PCB) comprising an electrowetting grid of electrodes forming a droplet pathway, an array of detection electrodes accessible to the droplet pathway, each comprising a self-assembled monolayer and a capture probe, and a plurality of interconnections from the electrowetting grid and the detection electrodes. The top plate comprises a conductive surface substantially parallel to the bottom substrate and mated thereto to form a reaction chamber. In a further aspect, the bottom substrate further comprises a plurality of amplification pathways of electrowetting pads. In an additional aspect, some of the pads of the electrowetting grid comprise dried assay reagents. These can include, but are not limited to, deoxyribonucleotide triphosphates (dNTPs; usually a mixture of dCTP, dTTP, dGTP and dATP); sets of PCR primers, label probes, enzymes (reverse transcriptase (in the case where the target nucleic acid is RNA), exonucleases, polymerases (particularly heat stable enzymes such as Taq polymerase and variants thereof, as well as “Hot Start” embodiments).

[0005] In a further aspect, the array of detection electrodes in is fluid communication with the droplet pathway.

[0006] In a further aspect, the top plate can comprise fluid passageways spatially corresponding to the intended receiving pads of the electrowetting grid.

[0007] In an additional aspect, the cartridge further comprises a liquid reagent module (LRM) comprising a plurality of blisters comprising assay reagents, fluid passageways connecting each of said blisters to one of the fluid holes of the top plate, and a sample inlet port in fluid connection with the reaction chamber. In some aspects, the LRM further comprises an aliquot of capture beads, particularly magnetic capture beads. In an additional aspect, the fluid passageways of the LRM allow the assay reagents stored in the blisters to be dispensed at a location remote from the blister upon rupture of the blister. In a further aspect, the blisters of the LRM can

contain an immiscible fluid, particularly immiscible oil, lysis buffer, binding buffer and/or elution buffer.

[0008] In a further aspect, the biochip cartridge further comprises an external housing comprising a latched cover for irreversibly sealing the sample inlet port. In some aspects, the external housing further comprises electronic connections from the edge interconnectors of the bottom substrate and/or from the thermal zone connections. In an additional aspect, the external housing is asymmetrically shaped to facilitate only one insertion orientation into the bays of the devices herein. In a further aspect, the external housing can further comprise a barcode.

[0009] The present invention further provides methods of using the biochips of the invention. Thus, in one aspect, the invention provides methods of detecting a plurality of target nucleic acids in a sample comprising adding sample to the biochips of the invention, executing steps to lyse the cells of the sample, purify the sample, amplify the sample, and detect the sample, with optional washing steps at any or all operations.

[0010] In one aspect, the methods provide adding the sample to a biochip cartridge of the invention and executing assay operations comprising mixing the sample with lysis buffer, adding binding buffer and capture beads to the sample, mixing the beads and sample, optionally washing the beads, eluting the target nucleic acids from the beads, adding amplification reagents to the target nucleic acids to amplify the target nucleic acids to form amplicons, optionally digesting one strand of the amplicon using exonuclease, adding signaling probes to the amplicons to form hybridization complexes, binding the hybridization complexes to the capture probes on the detection electrodes to form assay complexes, optionally washing the detection electrodes, and electrochemically detecting the assay complexes.

[0011] In a further aspect, the invention provides an apparatus for the detection of target analytes comprising: a) an instrument bank comprising a plurality of biochip cartridge bays for insertion and analysis of a biochip cartridge; b) a touch screen display having a plurality of bay icons, each icon uniquely corresponding to one of the plurality of bays; wherein when a biochip cartridge is inserted into one of said bays the corresponding icon is enlarged and/or exhibited.

[0012] In an additional aspect, the invention provides an apparatus for the detection of target analytes comprising: a) an instrument bank comprising a plurality of biochip cartridge bays for insertion and analysis of a biochip cartridge; b) a touch screen display having a plurality of bay icons, each bay icon uniquely corresponding to one of the plurality of bays; wherein when one of said bay icons is touched a panel of first options about the corresponding bay is enlarged and/or exhibited.

[0013] In a further aspect the plurality of biochip cartridge bays are arranged in at least one vertically disclosed bank of bays, and the bay icons are similarly displayed. Similarly, the plurality of biochip cartridge bays can be arranged in at least two vertically disclosed banks of bays, and the bay icons are similarly displayed. Additionally, the plurality of biochip cartridge bays can be arranged in at least three vertically disclosed banks of bays, and the bay icons are similarly displayed. Similarly, the plurality of biochip cartridge bays can be arranged in at least four vertically disclosed banks of bays, and the bay icons are similarly displayed.

[0014] In an additional aspect, the panel of first options comprises a plurality of secondary icons each selected from