

detection chambers including reconstituted reagents such as blocking agents, buffers, labeled binding reagents and/or control/calibration reagents picked up in pill zones comprised in sample conduit branches 1440 and/or 1441); ii) introducing a liquid reagent from reagent chamber 1425 into detection chambers 1445 and/or 1446 (optionally, the reagent introduced into the detection chambers including reconstituted reagents such as blocking agents, buffers, labeled binding reagents and/or control/calibration reagents picked up in pill zones comprised in reagent conduit 1470); iii) washing detection chambers with a wash reagent contained in reagent chamber 1426 (the reagent preferably comprising an electrochemiluminescence coreactant and providing a suitable environment for an ECL measurement) and iv) interrogating the contents of the detection chamber (preferably, by conducting an ECL measurement). Optionally, a wash step is included between steps (i) and (ii). Advantageously, the use of a two step format in binding assays allow analyte or other components in a sample to be bound to immobilized binding reagents in the detection chambers and washed out of the detection chamber prior to the introduction of labeled detection reagents (e.g., labeled binding reagents for use in sandwich binding assays or labeled analytes for use in competitive assays); carrying out assays in two steps may be advantageous in competitive assays and assays that suffer from large sample matrix effects or hook effects. Some assays may not require a wash step (e.g., non-washed ECL assays may be carried out by incorporating adding an ECL coreactant to the sample); for cartridges carrying out such non-washed assays (in one or two step formats), reagent chamber 1426 may be omitted.

[0274] As shown in FIG. 14b, a preferred embodiment of cartridge 1400 uses a laminar cartridge design employing a two part cartridge body (1410 and 1411) and cover layers 1401, 1402, 1403 and 1407. To allow for adequate sample and/or reagent volumes, the cartridge body has a thicker portion which includes features (channels, grooves, wells, compartments, etc.) that define, in part, the sample, reagent and waste chambers. The remainder of the cartridge is, preferably, much thinner so as to minimize cartridge weight, volume and material costs. The two part cartridge design is not required but is advantageous for producing the cartridge by low cost injection molding techniques by allowing the thicker regions of the cartridge body to be hollowed out thus reducing the amount of material needed to produce a cartridge, reducing the time required to cool the parts before ejection from an injection mold die and reducing the part deformation after release from the mold. In this hollowed out design, through-holes through the cartridge body can be provided for by tubes incorporated into body components 1410 and/or 1411 (see, e.g., tube 1439 in FIG. 14b). These tubes may be mated to tubes or holes in the other body component to form through-holes through the body. This mating can be accomplished by a variety of methods including tube mating methods known in the art. Preferred techniques include plastic welding techniques and/or the use of press fits (preferably, by mating a tapered tube with an outer diameter that decreases from d_{max} to d_{min} at its end with a tube that has an inner diameter between d_{max} and d_{min}). In an alternate embodiment, a one part cartridge body is used.

[0275] At least portions of the sample, reagent and vent conduits are formed by sealing cover 1403 on lower cartridge body part 1410. Detection chambers 1445 and 1446, portions of sample conduit branches 1440 and 1441 and portions of

elongated reagent conduit 1470 are formed by sealing cover layer 1407 (having patterned conductive layer 1423 (which forms a patterned electrode array analogous to the electrode array 963, shown in FIG. 9) and patterned dielectric overlayers 1421, 1422) to lower cartridge body part 1410 through intervening gasket layer 1405 (preferably, made from double sided adhesive tape). The detection chamber's depth, length and width are defined by cutouts 1447 and 1448 within the gasket layer. Cutouts 1406, 1408, 1412, 1413 in the gasket layer expose regions of dielectric layers 1421 and 1422 to sample conduit branches 1440 and 1441 and elongated reagent conduit 1470. Advantageously, dry reagent pills comprised within these reagents are located on these regions. This choice of pill locations allows dry reagent pills and/or immobilized reagents within the detection chambers to be dispensed on a single substrate. Preferably, as shown in FIG. 14, sample conduit branches 1440 and 1441 have segments that are adjacent and/or substantially parallel to detection chambers 1445 and 1446 and a U-turn segment to allow connection to the detection chambers. This arrangement provides for conduit lengths that are long enough to allow for the introduction of a sample to the conduit and mixing of the sample with a pill in the conduit prior to introduction of the sample to the detection chamber. These lengths are achieved without adding to the length of the cartridge. Advantageously, this arrangement also allows the patterned electrode layer to be used to conduct capacitive or conductometric measurements of fluid within the sample conduits as described above. Similarly, elongated reagent conduit 1470 has entrance and return segments, connected via a U-turn segment that is parallel to detection chambers 1445 and 1446. Lower cartridge body component 1410 further includes electrical access regions 1432 and 1433 that, together with cutouts 1417 and 1418 in gasket layer 1405 allow electrical contact to be made with conductive layer 1423.

[0276] Cover layer 1402 mates to lower cartridge body component 1410 to define conduit segments 1805 (readily seen in FIG. 18a) that (by connecting two Z-transitions) act as bridge segments connecting the fluidic networks defined by cover layers 1403 and 1407. Optionally, pill zones formed on cover layer 1402 on surfaces of bridge segments comprised within the sample or reagent conduits may be used to introduce dry reagents to the sample or liquid reagents. Cover layer 1401 mates to upper cartridge body component 1411 and seals reagent chambers 1425 and 1426, preventing the release of fluid from ampoules within the chambers. Cover layer 1401 also seals top side conduit segments including double Z-transition connecting segments such as segments 1810 and 1815 readily seen in FIG. 18a.

[0277] FIG. 15a shows a top view of upper body component 1411. FIGS. 16a and 16b show top and bottom views of lower body component 1410. As shown in FIG. 15a, the upper cartridge component 1411 preferably includes reagent chambers 1425, 1426 that are configured to hold reagent ampoules. Filters 1515, 1516 are preferably integrally molded into the upper cartridge component to ensure that substantially all of the glass fragments from the ruptured glass ampoules are not permitted to enter the fluidic network and possibly obstruct/block fluid flow. Alternatively, the filters may be separate components that are incorporated into the sample and/or assay reagent chambers during the manufacturing/assembly process; e.g., inserts that may preferably be snapped into place (see, e.g., inserts 2020 and 2021 in FIG. 20).