

biological analyte, if any, detected in the second measurement will include biological analyte from live microbial cells in the sample.

**[0183]** Methods to detect the presence of a microorganism in a sample can include the use of the detection devices disclosed herein. In certain embodiments, the method comprises providing i) a sample suspected of containing cells, ii) a detection article comprising a housing with two or more receptacles and an opening configured to receive the sample, iii) a cell concentration agent, and iv) a means for isolating and transferring the cell concentration agent from a upper receptacle to a lower receptacle in the housing, and v) a hydrogel comprising a cell extractant. In these embodiments, the detection device can comprise any one of the detection devices **100**, **200**, **300**, or **400**, shown in FIGS. **1-4**. Optionally, the detection device can comprise the cell concentration agent and/or the hydrogel.

**[0184]** The method further comprises transferring the sample into an upper receptacle in the housing wherein, in a liquid medium, the sample material is contacted with the cell concentration agent. The sample can comprise liquids, solids, semi-solids, or combinations thereof, which are transferred into the upper receptacle of the housing. If the sample does not comprise a liquid medium, a liquid medium (e.g., water or a buffered solution) can be added to the upper receptacle. A cell concentration agent is added to the liquid sample. The cell concentration agent is allowed to contact the liquid sample for a period of time. Optionally, the mixture can be mixed during the contact period by, for example, shaking, stirring, vortexing, and/or vibrating the housing. Preferably, the housing is closed (e.g., with optional cap) during the contact period to avoid loss of the sample and/or cell concentration agent.

**[0185]** The method further comprises isolating, from at least a portion of the liquid medium, the cell concentration agent, wherein isolating the cell concentration agent comprises transferring the cell concentration agent to a lower receptacle in the housing. As described herein, there are a variety of means for isolating the cell concentration agent. Non-limiting examples of means to isolate and transfer the cell concentration agent include partitioning and transferring the cell concentration agent through a passageway using a plunger (see FIGS. **1** and **2**), collecting and transferring the cell concentration agent in the cavity of a one-way valve (see FIG. **3**), and concentrating and transferring the cell concentration agent using a drain valve and a plunger (see FIG. **4**).

**[0186]** The method further comprises forming a liquid mixture comprising the isolated cell concentration agent and the hydrogel, wherein the cell extractant is released into the mixture. The liquid mixture comprising the cell concentration agent is contacted with a hydrogel comprising a cell extractant. The hydrogel (e.g., a hydrogel bead) can be contacted with the liquid mixture in the upper receptacle and/or lower receptacle of the housing. In some embodiments, the lower receptacle of the housing contains the hydrogel (see FIGS. **1** and **3**) and the liquid mixture is contacted with the hydrogel when the mixture is transferred into the lower receptacle. In some embodiments, the hydrogel is disposed in a third receptacle (see FIGS. **2** and **4**), through which the liquid sample passes (thereby contacting the liquid sample with the hydrogel) as the liquid sample is transferred from the upper receptacle to the lower receptacle.

**[0187]** It is recognized that, although FIGS. **2** and **4** show the use of a plunger to pierce the frangible seals and transfer the cell concentration agent to the lower receptacle, alterna-

tive instruments (e.g., a swab, a pipette, a filter) could be used instead of a plunger. In a method where such alternative instruments are used, it is preferable to remove at least a portion of the liquid sample (e.g., by decanting, pipetting, filtering, or by opening the drain valve, if present) such that the entire liquid sample is not transferred to the second receptacle when the frangible seal is pierced by the alternative instrument.

**[0188]** The method further comprises detecting a biological analyte. The biological analyte can be detected, as described herein, in the lower receptacle of the detection device before an effective amount of cell extractant is released from the hydrogel into the liquid mixture comprising the cell concentration agent. The biological analyte can be detected, as described herein, in the lower receptacle of the detection device after an effective amount of cell extractant is released from the hydrogel into the liquid mixture comprising the cell concentration agent. The biological analyte can be detected, as described herein, in the lower receptacle of the detection device before and after an effective amount of cell extractant is released from the hydrogel into the liquid mixture comprising the cell concentration agent.

**[0189]** It is anticipated that any of the methods disclosed herein can further comprise a biological growth step. The growth step is facilitated by providing a nutrient medium to support the growth of a microorganism. The nutrient medium can be mixed with the sample before, during, or after the concentration of microorganisms by the cell concentration agent. In some embodiments, the biological growth step occurs after the microorganisms have been concentrated by the cell concentration agent but before the biological analyte is detected. In some embodiments, the nutrient medium can contain nutrients and/or selective agents (e.g., salts, antibiotics) that favor the growth of certain types of microorganisms over other microorganisms that may be present in the sample.

Method of Concentrating a Particulate Cell Concentration Agent:

**[0190]** The present disclosure provides devices for concentrating a particulate cell concentration agent. The method includes providing a device to separate a portion of a liquid sample from a suspension the particulate material in the liquid sample. Suitable devices include, for example, the devices shown and described in FIG. **2A**, FIG. **3A**, FIG. **4A**, FIG. **5A**, FIG. **7A**, and FIG. **10A**. The devices each comprise a housing to contain a liquid sample including a particulate cell concentration agent and a means for separating the particulate cell concentration agent from at least a portion of the liquid sample.

**[0191]** In FIG. **2A**, the means for separating the particulate cell concentration agent includes the taper region **218** and the plunger comprising a lower seal **256**. In FIG. **3A**, the means for separating the particulate cell concentration agent includes the dead-end valve **370** and valve actuator **372**. In FIG. **4A**, the means for separating the particulate cell concentration agent includes the drain valve **480** and valve gate **482**. In FIGS. **5A** and **7A**, the means for separating the particulate cell concentration agent includes the plunger comprising fluid path with a filter **596** disposed in the fluid path. In FIG. **10A**, the means for separating the particulate cell concentration agent includes the plunger with a scraper that is configured to permit the passage of liquid between the edge of the scraper and the housing.