

is at least about 10  $\mu\text{m}$  or larger. As larger pore size filters are utilized, the sample will be easier and quicker to filter as the back pressure decreases with increase in pore size.

**[0165]** Filtering the sample can be accomplished using known methods. In an embodiment, the method of filtering that is chosen can be dictated at least in part on the particular application of the method. For example, the sample can be filtered using a negative vacuum, by applying a positive pressure, by the force of gravity. The particular technique used to filter the sample can depend at least in part on the type of device that is being utilized to carry out the method. For example, in order to utilize a negative vacuum, the device can be configured with a port that can be or reversibly attached to a source of vacuum; and in order to apply a positive pressure, the device can be configured to allow a user to apply a positive pressure by applying a force with their hands. In an embodiment, the sample can be filtered by applying a positive pressure.

**[0166]** Filtering using positive pressure (or using the force of gravity) can offer the advantage of easily being able to carry out the method in the field without the need for any further equipment, such as a vacuum pump.

**[0167]** In some embodiments, a centrifugation step may include the use of a relatively low-speed centrifugation in which the cell concentration agents separate (e.g., by sedimentation) out of the liquid but microorganisms (e.g., bacteria, yeast molds, spore) that are not bound to the cell concentration agent remain suspended in the liquid.

**[0168]** Optionally, the cell concentration agent can be resuspended in a wash solution (e.g., water or a buffer solution) and the cell concentration agent can be isolated from at least a portion of the wash solution. It will be recognized that a washing step can function to remove from the liquid sample contaminating materials that may interfere with a growth and/or detection process.

**[0169]** The method further comprises forming a liquid mixture comprising the isolated cell concentration agent and the hydrogel, wherein the cell extractant is released from the hydrogel. In some embodiments, when the cell concentration agent is isolated from at least a portion of the liquid sample, the cell concentration agent remains in a residual volume of liquid. Additional liquid (e.g., water or a buffer solution) optionally can be added to the cell concentration agent. In the embodiments wherein the cell concentration agent is filtered out of the liquid sample, the cell concentration agent can be resuspended in a volume of liquid (e.g., water or a buffer solution). The liquid suspension comprising the cell concentration agent is contacted with the hydrogel, thereby releasing the cell extractant into the liquid mixture. In methods involving the use of filters to collect the cell concentration agent, the liquid suspension comprising the cell concentration agent can also comprise the filter. An effective amount of cell extractant can be released from the hydrogel to effect the release of biological analytes from cells, if present, in the mixture. The release of an effective amount of cell extractant can occur over a period of time (e.g., up to several seconds, up to several minutes, up to an hour, or longer).

**[0170]** The method further comprises detecting an analyte. The detection of the biological analytes can involve the use of a detection system. Detection systems for certain biological analytes such as a nucleotide (e.g., ATP, NADH, NAD), a polynucleotide (e.g., DNA or RNA) or an enzyme (e.g., NADH dehydrogenase or adenylate kinase) are known in the art and can be used according to the present disclosure. Meth-

ods of the present disclosure include known detection systems for detecting a biological analyte. Preferably, the accuracy and sensitivity of the detection system is not significantly reduced by the cell extractant. More preferably, the detection system comprises a homogeneous assay.

**[0171]** In some embodiments, detecting the biological analyte can comprise detecting the analyte directly in a vessel (e.g., a tube, a multi-well plate, and the detection devices described herein) in which the liquid mixture comprising the sample and the hydrogel comprising a cell extractant is formed. In some embodiments, detecting the biological analyte can comprise transferring at least a portion of the liquid mixture to a container other than the vessel in which the liquid mixture comprising the sample and the hydrogel comprising a cell extractant is formed. In some embodiments, detecting the biological analyte may comprise one or more sample preparation processes, such as pH adjustment, dilution, filtration, centrifugation, extraction, and the like.

**[0172]** In some embodiments, the detection system comprises a detection reagent. Detection reagents include, for example, dyes, enzymes, enzyme substrates, binding partners (e.g., an antibody, a monoclonal antibody, a lectin, a receptor), labeled binding partners, and/or cofactors. In some embodiments, the detection reagent comprises a hydrogel, such as the hydrogels comprising an enzyme or enzyme substrate, as described in U.S. Patent Application No. 61/101,546, filed on Sep. 30, 2008, and entitled "BIODETECTION ARTICLES". In some embodiments, the detection system comprises an instrument. Nonlimiting examples of detection instruments include a spectrophotometer, a luminometer, a plate reader, a thermocycler, an incubator.

**[0173]** Detection systems can include detection instruments. Detection instruments are known in the art and can be used to detect biological analytes colorimetrically (i.e., by the absorbance and/or scattering of light), fluorescently, or luminescently. Examples of the detection of biomolecules by luminescence are described by F. Gorus and E. Schram (Applications of bio- and chemiluminescence in the clinical laboratory, 1979, Clin. Chem. 25:512-519).

**[0174]** An example of a biological analyte detection system is an ATP detection system. The ATP detection system can comprise an enzyme (e.g., luciferase) and an enzyme substrate (e.g., luciferin). The ATP detection system can further comprise a luminometer. In some embodiments, the luminometer can comprise a bench top luminometer such as, for example, the FB-12 single tube luminometer (Berthold Detection Systems USA, Oak Ridge, Tenn.). In some embodiments, the luminometer can comprise a handheld luminometer such as, for example, the NG Luminometer, UNG2 (3M Company, St. Paul, Minn.).

**[0175]** In some embodiments, the biological analyte is detected at a single time point. In some embodiments, the biological analyte is detected at two or more time points. When the biological analyte is detected at two or more time points, the amount of biological analyte detected at a first time (e.g., before an effective amount of cell extractant is released from a hydrogel to effect the release of biological analytes from live cells in at least a portion of the sample) point can be compared to the amount of biological analyte detected at a second time point (e.g., after an effective amount of cell extractant is released from a hydrogel to effect the release of biological analytes from live cells in at least a portion of the sample). In some embodiments, the measurement of the biological analyte at one or more time points is performed by an