

instrument with a processor. In certain preferred embodiments, comparing the amount of biological analyte at a first time point with the amount of biological analyte at a second time point is performed by the processor.

[0176] For example, the operator measures the amount of biological analyte in the sample after the liquid mixture including the sample and the hydrogel comprising a cell extractant is formed. The amount of biological analyte in this first measurement (T_1) can indicate the presence of “free” (i.e. acellular) biological analyte and/or biological analyte from nonviable cells in the sample. In some embodiments, the first measurement can be made immediately (e.g., about 1 second) after the liquid mixture including the sample and the hydrogel comprising a cell extractant is formed. In some embodiments, the first measurement can be at least about 5 seconds, at least about 10 seconds, at least about 20 seconds, at least about 30 seconds, at least about 40 seconds, at least about 60 seconds, at least about 80 seconds, at least about 100 seconds, at least about 120 seconds, at least about 150 seconds, at least about 180 seconds, at least about 240 seconds, at least about 5 minutes, at least about 10 minutes, at least about 20 minutes after the liquid mixture including the sample and the hydrogel comprising a cell extractant is formed. These times are exemplary and include only the time up to that the detection of a biological analyte is initiated. Initiating the detection of a biological analyte may include diluting the sample and/or adding a reagent to inhibit the activity of the cell extractant. It will be recognized that certain detection systems (e.g., nucleic acid amplification or ELISA) can generally take several minutes to several hours to complete.

[0177] The operator allows the sample to contact the hydrogel comprising the cell extractant for a period of time after the first measurement of biological analyte has been made. After the sample has contacted the hydrogel for a period of time, a second measurement of the biological analyte is made. In some embodiments, the second measurement can be made up to about 0.5 seconds, up to about 1 second, up to about 5 seconds, up to about 10 seconds, up to about 20 seconds, up to about 30 seconds, up to about 40 seconds, up to about 60 seconds, up to about 90 seconds, up to about 120 seconds, up to about 180 seconds, about 300 seconds, at least about 10 minutes, at least about 20 minutes, at least about 60 minutes or longer after the first measurement of the biological analyte. These times are exemplary and include only the interval of time from which the first measurement for detecting the biological analyte is initiated and the time at which the second measurement for detecting the biological analyte is initiated. Initiating the detection of a biological analyte may include diluting the sample and/or adding a reagent to inhibit the activity of the cell extractant.

[0178] Preferably, the first measurement of a biological analyte is made about 1 seconds to about 240 seconds after the liquid mixture including the sample and the hydrogel comprising a cell extractant is formed and the second measurement, which is made after the first measurement, is made about 1.5 seconds to about 540 seconds after the liquid mixture is formed. More preferably, the first measurement of a biological analyte is made about 1 second to about 180 seconds after the liquid mixture is formed and the second measurement, which is made after the first measurement, is made about 1.5 seconds to about 120 seconds after the liquid mixture is formed. Most preferably, the first measurement of a biological analyte is made about 1 second to about 5 seconds after the liquid mixture is formed and the second measure-

ment, which is made after the first measurement, is made about 1.5 seconds to about 10 seconds after the liquid mixture is formed.

[0179] The operator compares the amount of a biological analyte detected in the first measurement to the amount of biological analyte detected in the second measurement. An increase in the amount of biological analyte detected in the second measurement is indicative of the presence of one or more live cells in the sample.

[0180] In certain methods, it may be desirable to detect the presence of live somatic cells (e.g., nonmicrobial cells). In these embodiments, the hydrogel comprises a cell extractant that selectively releases biological analytes from somatic cells. Nonlimiting examples of somatic cell extractants include nonionic detergents, such as non-ionic ethoxylated alkylphenols, including but not limited to the ethoxylated octylphenol Triton X-100 (TX-100) and other ethoxylated alkylphenols; betaine detergents, such as carboxypropylbetaine (CB-18), NP-40, TWEEN, Tergitol, Igepal, commercially available M-NRS (Celsis, Chicago, Ill.), M-PER (Pierce, Rockford, Ill.), CellLytic M (Sigma Aldrich). Cell extractants are preferably chosen not to inactivate the analyte and its detection reagents.

[0181] In certain methods, it may be desirable to detect the presence of live microbial cells. In these embodiments, the hydrogel can comprise a cell extractant that selectively releases biological analytes from microbial cells. Nonlimiting examples of microbial cell extractants include quaternary ammonium compounds, including benzalkonium chloride, benzethonium chloride, ‘cetrimide’ (a mixture of dodecyl-, tetradecyl- and hexadecyl-trimethylammonium bromide), cetylpyridium chloride; amines, such as triethylamine (TEA) and triethanolamine (TeolA); bis-Biguanides, including chlorhexidine, alexidine and polyhexamethylene biguanide dialkyl ammonium salts, including N-(n-dodecyl)-diethanolamine, antibiotics, such as polymyxin B (e.g., polymyxin B1 and polymyxin B2), polymyxin-beta-nonapeptide (PMBN); alkylglucoside or alkylthioglucoside, such as Octyl- β -D-1-thioglucopyranoside (see U.S. Pat. No. 6,174,704 herein incorporated by reference in its entirety); nonionic detergents, such as non-ionic ethoxylated alkylphenols, including but not limited to the ethoxylated octylphenol Triton X-100 (TX-100) and other ethoxylated alkylphenols; betaine detergents, such as carboxypropylbetaine (CB-18); and cationic, antibacterial, pore forming, membrane-active, and/or cell wall-active polymers, such as polylysine, nisin, magainin, melittin, phospholipase A_2 , phospholipase A_2 activating peptide (PLAP); bacteriophage; and the like. See e.g., Morbe et al., *Microbiol. Res.* (1997) vol. 152, pp. 385-394, and U.S. Pat. No. 4,303,752 disclosing ionic surface active compounds which are incorporated herein by reference in their entirety. Cell extractants are preferably chosen not to inactivate the biological analyte and/or a detection reagent used to detect the biological analyte.

[0182] In certain alternative methods to detect the presence of live microbial cells in a sample, the sample can be pre-treated with a somatic cell extractant for a period of time (e.g., the sample is contacted with a somatic cell extractant for a sufficient period of time to extract somatic cells before a liquid mixture including the sample and a hydrogel comprising a microbial cell extractant is formed). In the alternative embodiment, the amount of biological analyte detected at the first measurement will include any biological analyte that was released by the somatic cells and the amount of additional