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APPARATUS FOR POINT-OF-CARE DETECTION OF NUCLEIC ACID IN A SAMPLE

CROSS REFERENCE TO RELATED APPLICATIONS

This application is related and claims priority to U.S. Provisional Application entitled "Disposable Sample Preparation Cartridge" Ser. No. 61/348,169, filed on May 25, 2010, U.S. Provisional Application entitled "Polymeric Method for Solution Sequestration and Release" Ser. No. 61/348,155, filed on May 25, 2010, and to U.S. Provisional Application entitled "Disposable, Inexpensive Heater for Point of Care Diagnostics" Ser. No. 61/348,160, filed on May 25, 2010, the disclosure of each of which is incorporated herein by reference in its entirety. The present application is also related to U.S. Patent Application entitled "Methods for Point-Of-Care Detection Of Nucleic Acid In A Sample" Ser. No. 13/115,878, filed on May 25, 2011, herein incorporated by reference in its entirety.

STATEMENT OF GOVERNMENT GRANT

The United States Government has rights in this invention pursuant to Contract No. DE-AC52-07NA27344 between the United States Department of Energy and Lawrence Livermore National Security, LLC for the operation of Lawrence Livermore National Laboratory.

TECHNICAL FIELD

The present disclosure relates to methods and apparatus for detecting a target nucleic acid in a sample such as a biological sample or an environmental sample, and related methods and apparatus of point-of-care detection and diagnosis of a pathogen or condition in a subject.

BACKGROUND

Early detection of epidemic outbreaks impacting public health and/or veterinary medicine needs cost-effective, robust and specific assays. Due to these needs, such assays traditionally have been conducted in centralized laboratories, rather than at the point-of-care.

However, Point-Of-Care Testing promises to bring the test to the test subject, in either the field or the clinic, providing more rapid detection with potential benefit to both the test subject and to public health.

In order to facilitate Point-Of-Care Testing, assays should be run without access to large, fragile or expensive equipment commonly found in centralized laboratories. Such equipment may include centrifuges, vortexers, thermocyclers, microscopes and incubators. Furthermore, external power sources may be unavailable in the field, and specialized technicians may not be available to conduct assays. Indeed, in some circumstances it may be desirable for test subjects to conduct self-testing.

A few companies have successfully designed high specificity Point-Of-Care Testing equipment relevant to the detection of epidemic outbreaks. To achieve high specificity, this equipment typically relies on Polymerase Chain Reaction (PCR), conducted in the field, to confirm the presence of a suspected pathogen or agent. The Bioseq device from Smiths (Herts, U.K.) is one such hand-held instrument that can accurately detect bacterial and viral agents. Idaho's Razor

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(Salt Lake City, Utah) and Selex's Nexsense B (Edinburgh, U.K.) are two other commercially available systems.

Although PCR systems provide excellent specificity, they also greatly increase costs as compared to other less specific assay kits, such as an Enzyme-Linked Immunosorbent Assay (ELISA). Furthermore, PCR systems need very clean samples, necessitating large upfront investment in sample preparation platforms that effectively integrate with microfluidic sample handling and readout. As an example of PCR-based Point-Of-Care Testing capital expenditures, the Bioseq device has a base unit price in excess of \$10,000 (the base unit is used in conjunction with consumable sample insert cartridges).

In order to provide high specificity Point-Of-Care Testing equipment without necessitating formidable upfront investment by the end user, radical simplification of the processes used by current PCR-based Point-Of-Care Testing equipment is needed. Specifically, there is a need for affordable, rapid, specific and accurate Point-Of-Care Testing assays that are completely disposable, or that need only nominal upfront investment. In order to accomplish this, complex interactions at the interface of biology, chemistry and material science should be harnessed synergistically (FIG. 1).

SUMMARY

Provided herein are methods and apparatus for detecting a target nucleic acid in a sample, and related methods and apparatus for diagnosing a condition in an individual. In particular, the methods and apparatus herein described allow in several embodiments integrated sample acquisition, nucleic acid extraction, amplification and detection all in one tube, which allow rapid, point-of-care detection of nucleic acid and/or diagnosis of a condition that is associated with presence of a particular nucleic acid in an individual.

According to a first aspect of the current disclosure, apparatus for detecting a target nucleic acid in a sample is described. The apparatus comprises a containment vessel, a sample collection element configured for removable coupling to the containment vessel, a nucleic acid binding element positioned within the containment vessel. The sample collection element is configured to collect the sample and to transfer the sample to the nucleic acid binding element when the sample collection element is removably coupled to the containment vessel. The apparatus further comprises a plurality of reagents configured for placement in fluid communication with the nucleic acid binding element. The plurality of reagents comprises nucleic acid purification reagents and nucleic acid amplification reagents. The apparatus further comprises a heater configured to heat the nucleic acid amplification reagents in fluid communication with the nucleic acid binding element.

According to a second aspect of the current disclosure, apparatus for diagnosing a condition in an individual is described. The condition is associated to presence of a target nucleic acid in the individual, which is produced by certain pathogens. The apparatus comprises a containment vessel, a sample collection element configured for removable coupling to the containment vessel and optionally comprising a lumen, a nucleic acid binding element positioned within the containment vessel, a waste collection unit, a plurality of reagents, which may be enveloped within a plurality of reagent cartridges that are connected to the containment vessel, and a heater configured to heat the nucleic acid amplification reagents when the nucleic acid amplification reagents are in fluid communication with the nucleic acid binding element.