

According to a third aspect of the current disclosure, methods for detecting a target nucleic acid in a sample are described. The method comprises sealing the sample within a containment vessel, transferring the sample to a nucleic acid binding element position in the containment vessel, amplifying the target nucleic acid in the containment vessel, and detecting amplification of the target nucleic acid in the containment vessel.

According to a fourth aspect of the current disclosure, a method 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 method comprises sealing a sample from the individual within a containment vessel, transferring the sample to a nucleic acid binding element position in the containment vessel, purifying nucleic acid from the sample in the containment vessel, amplifying the target nucleic acid in the containment vessel, detecting amplification of the target nucleic acid in the containment vessel, and diagnosing the condition.

The methods and apparatus herein described allow in some embodiments inexpensive, rapid detection of a pathogen present in a sample, such as a biological sample collected from a subject or an environmental sample collected from a surface, soil, water or air and also inexpensive, point-of-care diagnosis of a condition in an individual, such as mammals and in particular human beings.

The methods and apparatus herein described can be used in connection with applications wherein detection of a pathogen is desired, including but not limited to medical application, biological analysis and diagnostics including but not limited to clinical applications.

The details of one or more embodiments of the disclosure are set forth in the accompanying drawings and the description below. Other features, objects, and advantages will be apparent from the description and drawings, and from the claims.

BRIEF DESCRIPTION OF THE DRAWINGS

The accompanying drawings, which are incorporated into and constitute a part of this specification, illustrate one or more embodiments of the present disclosure and, together with the detailed description and examples sections, serve to explain the principles and implementations of the disclosure.

FIG. 1 shows Venn Diagrams indicating designated sweet spots of disposable point-of-care diagnostics.

FIG. 2 shows Foot-and-mouth disease (FMD) is endemic in many parts of the world. In this map, black indicates countries where FMDv outbreaks have been reported between 1997-2008. Over the past few years, developing countries, including Sub-Saharan Africa, northern South America, Southeast Asia and the Middle East, have reported the most cases of FMD. (Data from U.N. Food and Agriculture Organization; U.N. Food and Agriculture Organization.)

FIG. 3A shows a schematic rendering of a disposable device according to several embodiments of a first structure.

FIG. 3B shows a photograph of an electronic heater according to several embodiments.

FIG. 4A shows a procedural flow chart of a method for detecting a target nucleic acid in a sample.

FIG. 4B shows a procedural flow chart of a method for diagnosing a condition in an individual.

FIG. 4C shows a procedural flow chart of a method for integrated sample acquisition, preparation, nucleic acid extraction, amplification and detection all in one tube using the apparatus according to the present disclosure.

FIG. 5A shows a schematic rendering of a disposable device according to several embodiments of a second structure.

FIG. 5B shows a schematic rendering of a disposable device according to several embodiments of a third structure.

FIG. 5C shows a schematic rendering of a disposable device according to several embodiments a fourth structure.

FIG. 6A shows a reagent cartridge according to several embodiments.

FIG. 6B shows a waste collection unit according to several embodiments.

FIG. 7 shows sequestration of nucleic acid amplification master mix and enzyme in separate PCL shells and temperature-based release of the shell contents according to several embodiments.

FIG. 8 shows images showing colorimetric detection of recombinant FMDv template: 3 negative, no template controls (-) and 3 positive template (+) samples are shown. (-) tubes remain violet while (+) tubes shift to blue.

FIG. 9 shows images of live FMDv dilution series from tissue homogenate run in prototype containment tubes. Top panel indicates starting reaction colors, and lower panel indicates reaction mix colors at 60 min. While material in all tubes amplified, reaction mixture colorimetric results show a violet purple to blue transition with increasing virus concentration.

FIG. 10 shows colorimetric detection of recombinant FMDv after 30 min.

FIGS. 11A-B show time sequence gel electrophoresis results from samples run in prototype reaction tubes.

FIG. 12 shows time series images of initial MRSA assay in device tubes according to several embodiments. The swabs were dipped in 100 μ L of 10 pg/ μ L DNA.

FIG. 13 shows time series images of MRSA assay in micro-centrifuge tubes. (-) represents no template controls. Numbers indicate pg DNA per reaction. Most dilute MRSA genomic DNA samples on right contain ~17 copies of DNA.

DETAILED DESCRIPTION

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 disclosure, apparatus for detecting a target nucleic acid in a sample is described.

The term "apparatus" as used herein indicates a device, a machine, an instrument or a system that performs certain function according to the current disclosure.

The terms "detect" or "detection" as used herein indicates the determination of the existence, presence or fact of a target in a limited portion of space, including but not limited to a sample, a reaction mixture, a molecular complex and a substrate. The "detect" or "detection" as used herein can comprise determination of chemical and/or biological properties of the target, including but not limited to ability to interact, and in particular bind, other compounds, ability to activate another compound and additional properties identifiable by a skilled person upon reading of the present disclosure. The detection can be quantitative or qualitative. A detection is "quantitative" when it refers, relates to, or involves the measurement of quantity or amount of the target or signal (also referred as quantitation), which includes but is not limited to