**Title:** HIGH THROUGHPUT POINT OF CARE ASSAY SYSTEMS

**Abstract:** Systems, devices, kits and methods for performing point of care assays, in particular for performing a plurality of concurrent isothermal nucleic acid amplification assays in a point of care environment.
High Throughput Point of Care Assay Systems

Summary of the Invention

The present invention relates to systems, devices, kits and methods for performing point of care isothermal nucleic acid amplification assays, in particular for performing a plurality of concurrent isothermal nucleic acid amplification assays in a point of care environment. The invention further relates to modular systems for performing point of care investigations, in particular for performing a plurality of concurrent assays in a point of care environment.

Background to the Invention

High throughput screening has been used in the pharmaceutical industry as a way of screening numerous drug candidates in a short period of time. However, there remains a need in the art for a device which can be used at the point of care to process numerous samples in real-time to determine microbial and viral content. Described herein are systems, devices, kits and methods which utilise isothermal nucleic acid amplification assays and/or other assays for the rapid identification of the microbial and viral content of samples, including clinical and environmental samples.

Summary of the Invention

Accordingly, in a first aspect the present invention provides a point of care system for performing a plurality of concurrent isothermal nucleic acid amplification assays on target sequences. The system comprises a plurality of ports each configured to receive a reaction vessel configured to perform an isothermal nucleic acid amplification assay on a target sequence in sample when received in said port. The system also includes at least one identification device/reader for gathering information relating to the reaction vessel and/or the sample. Also present is at least one detector for gathering information relating to amplified nucleic acid in each reaction vessel, a central processing unit programmed to process the information gathered relating to amplified nucleic acid in each reaction vessel and the information gathered relating to the identity of the sample and/or reaction vessel; and a display interface in communication
with the central processing unit for presenting information related to the amplification of nucleic acid in each sample. The central processing unit is typically in communication with both the at least one identification device and the at least one detector.

Reaction vessels suitable for use in the invention will typically comprise one, two, three, four, five or more reaction chambers. Each reaction chamber will usually be suitable for performing a different isothermal nucleic acid amplification assay on a different target sequence. Usually, the same sample will be tested in each reaction chamber of a reaction vessel. Where multiple reaction chambers are present, the temperature required to perform a nucleic acid amplification assay on a sample will usually be the same for each chamber.

Systems according to the invention may have from about two to about fifty ports, preferably from about two to about ten ports, more preferably from about two to about five ports, four ports being particularly preferred. However, embodiments with more than fifty ports are also contemplated.

Such systems have been found to be particularly advantageous because they greatly reduce the time needed to screen i) a single sample for a plurality of target sequences; and ii) a plurality of samples for any number of target sequences. By providing a point of care system which reduces the time required to screen multiple samples for one or more target pathogen(s), the pathogen(s) which are present can be identified and, ultimately, treated significantly more quickly compared with known techniques. Likewise, because results can be provided so much more quickly than with known techniques and systems, real-time monitoring of pathogens can be provided. This can have a profound effect on outcomes.

The system will often be configured to receive and process a plurality of reaction vessels simultaneously. Typically, the reaction vessels will be processed independently from one another. That is to say, the processing of one reaction vessel by the system will not be reliant or dependent upon the progress of another reaction vessel. Naturally, it will be clear that if all of the available ports are occupied, then further reaction vessels cannot be processed until a port becomes available.
In most embodiments, the processing of samples within reaction vessels is controlled by the central processing unit, in particular the heating of the samples and reagents, and the detection of amplified nucleic acid. To yield rapid analysis of samples containing target nucleic acids for amplification, the central processing unit controls the processing of each of the reaction vessels independently as well as concurrently. The central processing unit can thereby coordinate the processing of reaction vessels and allow the high throughput screening of samples.

To achieve a high throughput of screening it is preferred that an individual reaction vessel is processed to yield a result in less than about 60 minutes, preferably in less than about 15 minutes, less than about 12 minutes, less than about 10 minutes, less than about 9 minutes, less than about 8 minutes, less than about 7 minutes, less than about 6 minutes, less than about 5 minutes.

Preferably, an individual reaction vessel is processed to yield a result in at least about 1 minute, at least about 2.5 minutes, at least about 5 minutes, at least about 7 minutes, at least about 8 minutes, at least about 9 minutes, at least about 10 minutes, at least about 12 minutes, at least about 15 minutes, at least about 20 minutes, at least about 30 minutes, at least about 40 minutes, at least about 50 minutes, at least about 60 minutes.

In embodiments, an individual reaction vessel is processed to yield a result in from about 1 minute to about 60 minutes, 5 minutes to about 30 minutes, preferably from about 7 minutes to about 15 minutes, more preferably from about 8 minutes to about 12 minutes. In embodiments, an individual reaction vessel is processed to yield a result in from preferably from about 1 minute to about 10 minutes, from about 1 minute to about 8 minutes, from about 1 minute to about 5 minutes, from about 1 minute to about 2.5 minutes, from about 2.5 minutes to about 5 minutes, from about 2.5 to about 8 minutes, from about 2.5 to about 10 minutes.

Different reaction vessels may require different processing times depending on their target, their specificity, the reagents, the sample quality, and such forth. Having identified the reaction vessel, the central processing unit will typically select a
predetermined set of conditions appropriate for the identified reaction vessel, e.g. heating to the sample and reagents within the reaction chamber of the reaction vessel to a predetermined temperature.

In embodiments, the amplification of the polynucleotides is performed isothermally. For example, the amplification may be performed without subjecting the polynucleotides to thermal cycling, e.g., by maintaining a temperature of the polynucleotides to within +/- 20°C, to within +/- 15°C, to within +/- 10°C, to within +/- 5°C, to within +/- 2.5°C, or at a substantially constant temperature during amplification.

In embodiments, the amplification of the polynucleotides is performed without subjecting the polynucleotides to a temperature sufficient to denature double stranded polynucleotides during the amplification. For example, the amplification of the polynucleotides may be performed without subjecting the polynucleotides to a temperature in excess of about 90°C, about 80°C, about 70°C, or about 60°C during amplification. In embodiments, the amplification of the polynucleotides is performed without subjecting the polynucleotides to conditions sufficient to denature double stranded polynucleotides during the amplification. For example, the amplification may be performed without subjecting the polynucleotides to physical, chemical, or thermal conditions sufficient to denature double stranded polynucleotides during amplification.

In embodiments, the amplification of the polynucleotides is performed without first subjecting the polynucleotides to a temperature sufficient to denature double stranded polynucleotides present in the sample. For example, the amplification of the polynucleotides may be performed without first subjecting the polynucleotides to a temperature in excess of about 90°C, about 80°C, about 70°C, about 60°C, or about 55°C. In embodiments, the polynucleotides and/or amplicons thereof are detected without first subjecting the polynucleotides to such excess temperatures. In embodiments, the amplification of the polynucleotides is performed without first subjecting the polynucleotides to conditions sufficient to denature double stranded polynucleotides present in the sample. For example, the amplification may be performed without first subjecting the polynucleotides to physical, chemical, or thermal
conditions sufficient to denature double stranded polynucleotides present in the sample.

Typically, the sample and reagent mixture will be heated to a temperature of from about 37 °C to about 85 °C, preferably from about 37 °C to about 60 °C. The specific conditions will depend upon the specific assay. When a NEAR assay(s) are employed in the methods, systems and kits of the invention, the sample and reagent mixture and/or reaction chamber will typically be heated to a temperature within the range of from about 50 °C to about 60 °C, preferably from about 55 °C to about 59 °C. Alternatively if a RPA assay(s) are employed then the sample and reagent mixture and/or the reaction chamber will be heated to a temperature from about 37 °C to about 42 °C.

In embodiments, the amplification is performed in a total time T beginning with a step of combining the sample with reagents sufficient to perform amplification of target polynucleotides in the sample and ending when amplification has proceeded by an amount sufficient to permit the qualitative or quantitative determination of the polynucleotides or amplicons thereof. In other embodiments, the total time T begins with a step of combining the sample with reagents sufficient to release the polynucleotides from microorganisms within the sample. For example, the microorganisms may be cells, e.g., bacterial, fungal, or yeast cells, and the reagents may be lysing reagents. In other embodiments, the total time T begins with the release of polynucleotides of at least some of the microorganisms in the sample, e.g., time T may begin with lysis of the microorganisms. In any of such embodiments, the total time T may be about 45 minutes or less, about 30 minutes or less, about 20 minutes or less, or about 15 minutes or less.

In embodiments, the amplification of the polynucleotides includes amplifying the polynucleotides by at least about 10^6 fold, at least about 10^7 fold, at least about 10^8 fold, at least about 10^9 fold, at least about 10^{10} fold, at least about 10^{11} fold, or at least about 10^{12} fold. Such amplification may be performed within the time T.

In embodiments, the reaction chamber is a chamber of a microfluidic cartridge. The cartridge may include reagents sufficient to perform the amplification of the
polynucleotides in the sample. The reagents may be in dry form within the cartridge prior to introduction of the sample to the cartridge.

The system includes means for identifying the reaction vessel and means for identifying the sample. Typically, a specific reaction vessel will contain reagents to perform a specific isothermal nucleic amplification or a specific number of isothermal nucleic acid amplifications in its one or more reaction chambers. As such, a specific reaction vessel will typically be used to identify a specific or specified number of target sequence(s). Accordingly, by identifying which reaction vessel is being used enables the system to interpret the results of the nucleic acid amplification appropriately and, in embodiments, for the central processing unit to control the processing of the reaction vessel in a predetermined manner.

Reaction vessel(s) may be identified to the device by coded information on each reaction vessel, comprising, for instance, a bar code, a smart chip, an RFID tag or a key. Suitable barcodes include linear bar code, a 2D bar code, a QR code, an alphanumerical code.

The device for gathering information relating to the identity of the sample and/or reaction vessel may be a coded information reader. The coded identification information may be acquired by direct contact between a coded information reader and the coded information. Alternatively, the coded identification information may be acquired by a contactless interaction between information reader and the coded information.

The coded information may include information directing the system as to how to process the reaction vessel. Alternatively, the system, typically the central processing unit, may be preprogrammed to process reaction vessels such that once a vessel is identified the system will process it appropriately. In such embodiments, as new tests are developed the system can be reprogrammed to accommodate the new tests. The coded information may also include information relating to expiry dates, lot numbers, batch numbers, and such like.
Typically, the means for identifying the reaction vessel and the means for identifying the sample comprise a coded information reader in communication with said central processing unit. The system may include more than one coded information reader. Separate coded information readers and associated coded information may be used to identify the sample and the reaction vessel respectively. The coded information reader may form an integral part of the port, alternatively it might be a separate component of the system or, when multiple coded information readers are present, a combination thereof. In a preferred arrangement each port has at least one dedicated coded information reader, preferably a dedicated coded information reader which is able to read coded information provided on a reaction vessel which is received in the port. An advantage of a coded information reader forming part of the port is that reading of the code is more easily automated and less error prone. In contrast, an advantage of the coded information reader forming a separate component is that it simplifies the construction of the system. In embodiments, coded information readers for identifying the reaction vessel are located in the port and a separate component is used to identify the sample. Typically, there will be one coded information reader configured to identify reaction vessels per port.

A port is understood to mean a fixture suitable for receiving and holding a reaction vessel. Typically, engagement of a reaction vessel with a port will cause the port to send a signal to the central processing unit identifying that a reaction vessel is present. Typically, when a reaction vessel is engaged with a port the central processing unit will identify a reaction vessel using a coded information reader. Typically, a reaction vessel and port will mechanically interlock with one another as the reaction vessel is received in the port.

Samples are tested for target sequences (i.e. target nucleic acid sequences). Typically, the sample is a biological sample, a patient sample, a veterinary sample, or an environmental sample.

In embodiments, target sequences may be amplified from many types of samples including, but not limited to samples containing spores, viruses, cells, nucleic acid from prokaryotes or eukaryotes, or any free nucleic acid. The sample(s) may be isolated from any material suspected of containing the target sequence.
For example, for animals, such as humans, the sample may comprise blood, bone marrow, mucus, lymph, hard tissues, liver, spleen, kidney, lung, or ovary, biopsies, sputum, saliva, tears, faeces, or urine. Or, the target sequence may be present in air, plant, soil, or other materials suspected of containing biological organisms.

Alternatively, target sequences may be present in samples that may also contain environmental and contaminants such as dust, pollen, and soot (for example, from diesel exhaust), or clinically relevant matrices such as urine, mucus, or saliva.

Target sequences may also be present in waste water, drinking water, air, milk, or other food.

Depending on the concentration of these contaminants, sample purification methods known to those of ordinary skill in the art may be required to remove inhibitors for successful amplification. Purification may, for example, involve the use of detergent lysates, sonication, vortexing with glass beads, or a French press. This purification could also result in concentration of the sample target. Samples may also be further purified, for example, by filtration, phenol extraction, chromatography, ion exchange, gel electrophoresis, or density dependent centrifugation.

In particular embodiments, the sample can be added directly to the reaction reagents, or pre-diluted and then added to the reaction mix, in either case without prior purification of target nucleic acid.

The system also includes a detector for gathering information relating to amplified nucleic acid in each reaction vessel. The detector may provide optical or fluorescence monitoring, electrochemical monitoring, or other monitoring of the contents of the reaction vessel. The detector will typically be arranged such that it can gather information relating to amplified nucleic acid in a reaction vessel whilst the reaction vessel is received within the port. Typically, there will be at least one detector per port. When multiple reaction chambers are present in a single reaction vessel, typically one
detector will be able to monitor all of the reaction chambers, although in other embodiments multiple detector(s) may be present in each port.

The system also includes a central processing unit programmed to process the information gathered relating to amplified nucleic acid(s) in each said identified reaction vessel. Typically, the central processing unit will be in wired and/or wireless connection with the detector such that information relating to amplified nucleic acid(s) may be transmitted to the central processing unit. The central processing unit may be present in a dedicated computer in communication with the remainder of the system or be built into the housing of the system, alternatively it might be a present in a non-dedicated computer, programmed with appropriate software, which is connected to the remainder of the system for the purpose of running the tests and processing the data thereby produced.

The system also includes, a user interface for presenting information related to the amplified nucleic acid from each sample. The user interface may be in the form of a monitor, a projector, a printer, or such forth, or combinations thereof.

It is envisaged that the system of the invention may take one of various forms. For instance, the entire system may be enclosed within in a single housing. Alternatively, each of the components of the system may be provided separately. Alternatively, the more than one port, means for identifying each reaction vessel, and detector may be provided in a first housing, whereas the central processing unit and, optionally, the user interface, will be provided in a second or more housing(s). Where the components of the system are provided in separate housings, they will typically be in wired and/or wireless communication with one another. In embodiments, the system comprises a plurality of devices each comprising at least one port and at least one detector and at least one means for identifying a reaction vessel, wherein each device is connected wirelessly and/or wired to one another or located on a docking station, and in communication with a central processing unit.

The system may also include one or more reaction vessels. In embodiments, the system includes at least one liquid transfer device that includes a housing having a pipette tip and a plunger assembly; and a reaction vessel, wherein the housing of the
liquid transfer device is configured to sealably engage with the reaction vessel. In some embodiments, the housing of the liquid transfer device can include a seal component configured to sealably engage with the reaction vessel. In some embodiments, the reaction vessel can include a seal component configured to sealably engage with the liquid transfer device. The systems can further include a sample reservoir, and the reaction chamber can optionally be configured to lockably engage with the sample reservoir. The sample reservoir will contain the sample to be tested and typically an elution buffer.

The liquid transfer device can be configured to lockably engage with the reaction vessel, e.g., without dispensing, prior to dispensing, and/or after dispensing a liquid sample. In some embodiments, the reaction vessel includes one or more components of a biological reaction.

Suitable reaction vessels and liquid transfer devices for use in the invention are disclosed in WO2013/041713 which is incorporated herein by reference.

Alternatively, the reaction vessels of the invention may be in form of a cassette. Typically, such a cassette might include a sample reservoir, one or more reaction chambers for performing isothermal nucleic acid amplifications, and a liquid transfer device for transferring a portion of the sample to be tested from the sample reservoir to the reaction chamber(s). Alternatively, the cassette may not include a sample chamber or liquid transfer device. In which case, the sample would be prepared before being placed within the cassette for testing.

The system will also typically comprise at least one additional station adapted to securely hold the sample reservoir.

In all embodiments, the additional station(s) and the ports can be adapted to heat or cool the contents of the sample reservoir and reaction chambers respectively. The ports can also be adapted to provide agitation of the reaction vessel or a portion thereof. Where a plurality of ports are present each will be able to heat or cool the contents of the reaction vessel independently of each other port. Thus, reaction vessels of the invention may be heated or cooled independently. The heating or
cooling will typically be controlled by the central processing unit. Typically, the heating or cooling provided will be determined by the identification of the reaction vessel.

The present invention also provides a reaction vessel for receipt and/or received in systems according to the invention, or for use in the kits according to the invention. The invention therefore provides the combination of one or more reaction vessels and a system according to the invention.

Reaction vessels for use in the kits, methods and systems of the invention typically include a seal, preferably a lockable seal, configured to retain the contents of the reaction vessel once a nucleic acid amplification assay has been performed. This is so that the reaction vessels can be handled without risk of the products of any nucleic acid amplification causing contamination or an infection risk. This is important in point of care applications.

In embodiments of the invention, the reaction vessel contains the reagents for a recombinase polymerase amplification (RPA), such as a recombinase, a single strand binding protein and a polymerase. The recombinase may be selected from T4 UvsX, T6 UvsX, or RecA. The DNA polymerase may be selected from the group consisting of E. coli DNA polymerase I Klenow fragment, B. stearothermophilus polymerase (Bst), B. subtilis Phi-29 polymerase, B. subtilis polymerase I (Bsu). The single strand binding protein is typically gp32.

The reagents for a recombinase polymerase amplification which typically also include a crowding agent, ATP (adenosine triphosphate) or an ATP analogue, dNTP(s), or T4 bacteriophage UvsY. Preferred crowding agents may be selected from the group comprising (preferably consisting of) polyethylene glycol (PEG), dextran, polyvinylalcohol (PVA), polyvinypyrrolidone (PVP) or Ficoll.

When present, the PEG is preferably PEG1450, PEG3000, PEG8000 or PEG10000. PEG will preferably have a molecular weight between about 15000 and about 20000.

When present, the dNTP(s) is/are preferably selected from the group consisting of dATP, dGTP, dCTP, and dTTP.
When present, the ATP or ATP analogue is typically selected from ATP, ATP-γ-S, ATB-β-S, ddATP or a combination thereof.

Alternatively, the reaction vessel may contain the reagents for a nicking and extension amplification reaction (NEAR). NEAR reagents typically comprise a nicking enzyme, a forward template nucleic acid, a reverse template nucleic acid, and a polymerase.

Reagents are typically in dried form or freeze dried form but may be in liquid form.

The system and kits of the invention can process NEAR and/or RPA isothermal nucleic acid amplification assays concurrently.

The present invention also provides a kit for performing concurrent isothermal nucleic acid amplification assays. Such kits will typically comprise a system according to the invention and a plurality of reaction vessels, each configured to perform at least one isothermal nucleic acid amplification assay. The kit will typically include a selection of different reaction vessels, each reaction vessel being capable of performing a specific isothermal nucleic acid amplification assay or specified number of different isothermal nucleic acid amplification assays. Reaction vessels will usually be labelled so that the system can identify them. Likewise, reaction vessels will typically be labelled so that a user can visually identify them. Suitable reaction vessels for use in the kits of the invention have been discussed earlier in this application.

The present invention also provides a method for identifying a target sequence in a sample using isothermal nucleic acid amplification comprising the steps of collecting and preparing a sample to be tested; selecting a plurality of separate reaction vessels, each reaction vessel being capable of performing at least one isothermal nucleic acid amplification assay in at least one reaction chamber; loading each reaction vessel with the sample; providing conditions suitable for performing isothermal nucleic acid amplifications in each reaction chamber (e.g. heating them to an appropriate temperature for initiating the isothermal nucleic acid amplification); gathering information relating to amplified nucleic acid in the reaction chambers; identifying any target sequences by processing the information gathered in a preprogrammed central
processing unit; and, optionally, displaying the results on a user display interface such as a computer monitor, printer or such forth.

Typically, each reaction chamber is capable of performing an isothermal nucleic acid amplification assay for a target sequence. As discussed previously, a reaction vessel may be able to perform a certain number of different isothermal nucleic acid amplification assays, for instance one, two, three, four or five different isothermal nucleic acid amplification assays.

In embodiments, the plurality of reaction vessels used in the method are selected from a group of reaction vessels containing the same or more reaction vessels than the selection, preferably a larger group of reaction vessels, and wherein each reaction vessel is capable of performing at least one isothermal nucleic acid amplification assay.

The larger group of reaction vessels may be capable of identifying a plurality of different target sequences, preferably a group of target sequences greater in size than the number of reaction vessels selected.

Preferably, isothermal nucleic acid amplification assays may be performed concurrently. Typically, isothermal nucleic acid amplification assays may be performed independently.

Alternatively, the present invention provides a point of care method for identifying the prevalence of a target pathogen in a selection of samples using isothermal nucleic acid amplification. The method will usually involve the steps of: collecting and preparing a plurality of samples to be tested; selecting a plurality of reaction vessels, each reaction vessel being capable of performing at least one isothermal nucleic acid amplification assay in at least one reaction chamber; loading each reaction vessel with a different sample; providing conditions suitable for performing isothermal nucleic acid amplification in each reaction chamber (e.g. heating them to an appropriate temperature for initiating the isothermal nucleic acid amplification); gathering information relating to amplified nucleic acid in the reaction chambers; and identifying
which of the samples the pathogen is present in by processing the information gathered using a preprogramed central processing unit.

In embodiments, each reaction vessel is capable of performing the same isothermal nucleic acid amplification assay(s). The reaction vessels are normally selected from a larger group of reaction vessels wherein a portion of reaction vessels making up said group are capable of performing different isothermal nucleic acid amplification assays.

The isothermal nucleic acid amplification assays are preferably performed concurrently and independently.

The present invention also envisages a device for high throughput screening using isothermal nucleic acid amplification assay(s) for the determination of microbial and viral content of a sample, typically at or close to the point of sampling. The device will typically comprise a control module; a plurality of process modules in operable communication with the control module; a user interface; a communication port and a data display.

Typically, each process module comprises a reaction vessel, a fluid handling manifold, and a detector.

As with other embodiments of the invention, the module may be configured to perform recombinase polymerase amplification of nucleic acid. Alternatively, the process module is configured to perform nicking and extension amplification reaction to amplify nucleic acid. Preferably, the process module is configured to amplify DNA within a sample without first thermally melting the DNA.

The present invention also envisages a method for performing concurrent isothermal nucleic acid amplification assays. The method will typically include the steps of: providing a first reaction vessel containing a first sample and a second reaction vessel containing a second sample, each reaction vessel being configured to perform an isothermal nucleic acid amplification assay; associating said reaction vessels with ports configured to gather information relating to amplified nucleic acid; identifying the reaction vessels and samples located within said reaction vessels; performing
concurrent isothermal nucleic acid amplifications on the samples whilst the reaction vessels are located in the ports; gathering information relating to amplified nucleic acid from each reaction vessel; processing the information gathered relating to amplified nucleic acid with a central processing unit; and presenting information related to the amplified nucleic acid from each sample on a display interface.

The present invention also provides a modular system for performing concurrent isothermal nucleic acid amplification assays on target sequences comprising: a first process module comprising a port configured to receive a reaction vessel, which is configured to perform an isothermal nucleic acid amplification assay on a target sequence contained in a sample when received in said port, and a first detector for gathering information relating to amplified nucleic acid in said first reaction vessel; at least one second process module comprising a second port configured to receive a second reaction vessel, which is configured to perform an isothermal nucleic acid amplification on target sequence contained in a sample when received in said second port, and a second detector for gathering information relating to amplified nucleic acid in said second reaction vessel; means for identifying each reaction vessel and sample; a control module comprising central processing unit configured to identify each reaction vessel and sample and to process information related to amplified nucleic acid in said identified reaction vessels; and a user interface for presenting said processed information related to the amplified nucleic acid from said samples; wherein the first process module, at least one second process module and control module detachably connect to one another.

Preferably, the first device, at least one second device, means for identifying each reaction vessel and sample and user interface all communicate with the control module. Preferably, the control module controls the progress of the isothermal nucleic acid amplifications within the reaction vessels. Preferably the isothermal nucleic acid amplifications are performed concurrently, preferably independently.

The invention further provides a modular device for performing multiple isothermal nucleic acid amplification assays, comprising: a control module, comprising: a central processing unit, and a user interface, a plurality of process modules in communication with the control module, each comprising a reaction chamber for performing an
isothermal nucleic acid amplification assay on a sample, and a detector for gathering information relating to amplified nucleic acid; and optionally, a data display module in communication with the control module; wherein, in use, the control module communicates operation commands to each of the plurality of process modules to initiate the isothermal nucleic acid amplification and the process modules each communicate information gathered in relation to amplified nucleic acid to the control module. Preferably the isothermal nucleic acid amplification assays may be performed concurrently, typically independently.

Typically, the control module is programmed to send and receive data to the plurality of process modules, said data relating to: mixing sample(s) with reagents once sample(s) introduced to reaction chamber; modulating the temperature of the reaction chamber to desired temperature; monitor status of plurality of reactions; actuate detector to acquire a measurement response to determine the presence or absence of target nucleic acid in the one or more sample(s); format measurement response data and output result on user interface; and/or communicate output results via communication port to a remote display terminal or a printer.

As with all other embodiments and aspects of the invention, the amplification of DNA may comprise recombinase polymerase amplification and/or nicking and extension amplification reaction.

The invention also provides a system interface for connecting and controlling a plurality of process modules for performing isothermal nucleic acid amplification assays comprising a backbone connector comprising n+1 terminals to engage n+1 isothermal nucleic acid amplification assay process modules; a user interface; a control module for sending and receiving data to the n+1 isothermal nucleic acid amplification assay modules via the user interface and backbone connector; a common power source for providing power to the network and the n+1 connected isothermal nucleic acid amplification assay modules.

In embodiments, the n+1 terminals of the backbone connector are specific to individual isothermal nucleic acid amplification assay modules. In other embodiments, the n+1
terminals of the backbone connector are non-specific and may be connected to more than one isothermal nucleic acid amplification assay module.

The control module typically manages the flow of data between the user interface and the n+1 isothermal nucleic acid amplification assay modules. Normally, the system may further comprise an identification reader, preferably a bar code scanner, a RFID scanner, a smart card reader, or a camera. Typically, the identification reader receives data relating to an individual from which a sample for analysis has been obtained. Alternatively or additionally, the identification reader may receive data relating to a reaction vessel for use in one of the process modules. Such data is typically used to configure the process module to perform a defined isothermal nucleic acid amplification analysis on a sample inserted in the process module.

In embodiments, n is an integer greater than 1, preferably between 1 and 49, more preferably between 1 and 9, more preferably between 1 and 4.

In a further aspect the invention provides a modular system for performing concurrent assays on at least one sample to investigate the biological content thereof.

The modular system may comprise at least one first process module configured to receive at least one first reaction vessel configured to perform at least a first assay on a sample when received in said at least one first process module. The system may also comprise at least one second process module configured to receive at least one second reaction vessel configured to perform at least a second assay on a sample when received in said at least one second process module. Optionally, the system may comprise at least one further process module configured to receive at least one further reaction vessel configured to perform at least a further assay on a sample when received in said optional further process module.

The system may comprise at least one identification device for gathering information relating to the identity of each reaction vessel and/or each sample.

The modular system may comprise at least one detector per process module for gathering information relating to the output of each assay in each reaction vessel; a
central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to provide information relating to the biological content of each sample; and a display interface in communication with the central processing unit which presents information relating to the biological content of each sample.

Typically, the first, second and optional further process modules are different. Typically, process modules are configured to receive different reaction vessels and/or perform different assays to one another.

Accordingly, the modular system can perform the same assay on a plurality of samples, or a plurality different assays on the same sample, or a plurality of different assays on a plurality of different samples, or combinations thereof. The assays may be performed independently and/or concurrently. Because the system is modular, the user is able to select an appropriate group of assays and process modules for the investigation they wish to perform, whilst only requiring the laboratory space typically associated with a single assay test device.

Such modular systems have been found to be particularly advantageous because they greatly reduce the time needed to screen i) a single sample for a plurality of target analytes; and ii) a plurality of samples for any number of analytes. By providing a point of care system which reduces the time and/or space required to screen multiple samples for one or more target analyte(s), the analyte(s) which are present can be identified more efficiently and, ultimately, treatment can be delivered more quickly compared with known techniques. Likewise, because results can be provided so much more quickly than with known techniques and systems, real-time monitoring of target analytes(s) can be provided. This can have a profound effect on outcomes.

Target analytes suitable for detection by the modular systems of the invention may include pathogens, biomarkers, drugs, drug metabolites, electrolytes, blood gases, (bio)chemicals [e.g. lipids, enzymes, hormones, sugars etc].
A process module may be a meter for a fluorescence immunoassay wherein the reaction vessel is selected from a plurality of assay cartridges. Suitable assay cartridges may be capable of testing for congestive heart failure, disseminated intravascular coagulation or thromboembolic events, myocardial infarction, or drug and/or drug metabolites, such as of paracetamol, amphetamines, methamphetamine, barbiturates, benzodiazepines, cocaine, methadone, opiates, phenycyclidine, THC and tricyclic antidepressants in urine. In particular, the assay cartridges may be suitable for the quantitative measurements of B-type natriuretic peptide (BNP) in EDTA anticoagulated whole blood or plasma; quantitative determination of cross-linked fibrin degradation products; quantitative determination of creatine kinase MB, myoglobin and troponin I; qualitative determination of the presence of drug and/or major metabolites above threshold concentrations. Suitable cartridges are sold under the brand name Alere Triage®.

Additionally, or alternatively, a process module may be a meter for performing reflectance photometry wherein the reaction vessel is a cassette for the quantitative determination of total cholesterol, HDL (high-density lipoprotein) cholesterol, triglycerides and/or glucose in whole blood. Cholesterol measurements are used in the diagnosis and treatment of disorders involving excess cholesterol in the blood and lipid and lipoprotein metabolism disorders. HDL (lipoprotein) measurements are used in the diagnosis and treatment of lipid disorders (such as diabetes mellitus), atherosclerosis, and various liver and renal diseases. Triglyceride measurements are used in the diagnosis and treatment of patients with diabetes mellitus, nephrosis, liver obstruction, other diseases involving lipid metabolism, or various endocrine disorders. Glucose measurements are used in the diagnosis and treatment of carbohydrate metabolism disorders including diabetes mellitus, neonatal hypoglycemia, and idiopathic hypoglycemia, and of pancreatic islet cell carcinoma. Suitable cassettes are sold under the brand name Alere LDX®.

Additionally, or alternatively, a process module may be for reading a reaction vessel in the form of an INR (International Normalized Range) test strip for managing anticoagulation therapy. Suitable test strips are sold under the brand name Alere INRatio®2 PT/INR.
Additionally, or alternatively, a process module may be for reading a reaction vessel in the form of a BGEM (blood gases, electrolytes, and metabolites) test card. Suitable test cards are available under the brand name Alere epoc®.

Additionally, or alternatively, a process module may be for reading a reaction vessel in the form of a blood glucose strip.

Additionally, or alternatively, a process module may be for reading a reaction vessel for performing a pregnancy test.

Additionally, or alternatively, a process module may be for reading a reaction vessel in the form of a urine test cartridge suitable for the quantitative determination of HbA1c and ACR (Albumin/Creatinine Ratio). Such process modules help identify patients at risk of complications of diabetes or hypertension. Suitable cartridges are available under the brand name Alere Afinion ®.

Additionally, or alternatively, a process module may be for reading a reaction vessel in the form of a cartridge for screening a plurality of drugs or drug metabolites in oral fluid. Such drugs or metabolites may be selected from the group consisting of amphetamine, methamphetamine, cocaine, cannabis, benzodiazepines, opiate, methadone, ketamine and buprenorphine. Suitable test cartridges are sold under the brand name Alere™ DDS®2 Test Cartridge ®.

Additionally, or alternatively, a process module may be for reading a reaction vessel for performing RPA or NEAR as described elsewhere in this application. Suitable test cartridges are sold under the brand name Alere i™

The modular system may comprise a control module comprising i) a housing configured to receive at least the at least one first and the at least one second process modules, and optionally to receive the optional further process module, within said housing, ii) the central processing unit, iii) the display interface, iv) optionally the identification device, and optionally iv) a power supply.
Typically, the process modules detachably connect to the control module. Additionally, or alternatively, the process modules are detachably receivable within the housing and/or slidably receivable within the housing. Typically, the process modules may detachably connect to the control module, but not to one another.

The housing may be configured to receive process modules in a vertically stacked arrangement.

The housing may be configured to receive from two to ten process modules, preferably from two to six process modules.

The housing may be configured to receive process modules of different heights. The housing may include a series of horizontal runners on an inner wall thereof for receiving the process modules. Typically, the runners are in the form of elongate protrusions and may be substantially equally spaced along at least the two inner walls of the housing. Typically, the at least two inner walls including the runners extend from a front of the housing including an opening for receiving the process modules to a rear of housing. The rear of the housing may be open or closed, typically closed.

The process modules may be of the same or different height. Typically, the first process module has a different height to the second process module and/or the third process module. Where process modules are of different height they may be configured to be received on the equally spaced runners. The process module may have a minimum height substantially equal to the vertical distance between adjacent runners within the housing. Process modules of a different height may have a height substantially equal to an integer multiple of the minimum height. Alternatively, the process modules may all be of the same height and configured for receipt on the equally spaced runners.

The process modules may have a height from about 20 mm to about 240 mm, or from about 30 mm to about 180 mm; or from about 40 mm to about 120 mm. The ratio of the height of the smallest process module to the largest process module may be from about 1:1 to about 1:12; or from about 1:1 to about 1:8; or from about 1:1 to about 1:4;
or from about 1:1 to about 1:3; or about 1:1; about 1:2; about 1:3; about 1:4; about 1:5; about 1:6; about 1:7 or about 1:8.

Alternatively, or additionally, the modular system may comprise a control module comprising the central processing unit, the display interface and optionally the at least one identification device, wherein the first and second process modules and the control module detachably connect to one another. In particular, the first process module may detachably connect to the second process module, and the control module may detachably connect with the first and/or second process module. If more than two process modules are present, all of the process modules may connect with one another. Likewise, the control module may detachably connect to each of the process modules. Typically, the control module will only connect to one process module at once.

The modular system may further comprise a power module for powering the process modules and the control module. Typically, the power module comprises a mains power supply and/or a battery. In the modular system the optional power module may detachably connect to any one of the process modules to provide power thereto. Typically, the means for connecting the process modules, control module and power module to one another will also provide the power connection between each such module.

The process modules, control module and, optional, power modules may be detachably connected in a vertical stack arrangement, preferably wherein the control module is located at a top of the stack, and the optional power module is locatable at a base of the stack. Typically, the control module will comprise physical connection means on its base. Typically, the power module will comprise physical connection means on its upper surface. Typically, the process modules will each comprise physical connection means both on their upper surface and their base.

The process modules are in wired or wireless communication with the control module. The wired communication may be by way of a ribbon-cable or by electronic connection means located on a surface of the process modules and/or control module. The electronic connection means may be located on an upper surface and/or the base of
the process modules. The electronic connection means may be located only on the base of the control module. Typically, the electronic connection means may be collocated with or in the physical connection means. The electronic connection means may provide power as well as wired communication. Alternatively, or additionally a power connection may be provided via the physical connection means.

In the modular system aspects of the invention the first and second reaction vessels may be different, and/or the first and second assays may be different.

The first and second assays may be different and selected from the list consisting of isothermal nucleic acid amplification assays, such as NEAR and RPA. The assays may be the same type but test for different pathogens or target analytes, or different types of assay testing for the same or different pathogens or target analytes. The reaction vessels may be configured to perform a plurality of different assays on the same and/or different samples.

The first, second or more assays may be a RPA assay for one or more of Flu, HIV, MRSA, streptococcal infection, Zika, Chlamydia, Gonorrhoea, Ebola, and the like. A combination of both viral and bacterial/fungal pathogens may be investigate simultaneously. A particular advantage when considering Flu, for example, is that from a serology perspective the flu strain can vary from outbreak to outbreak. RPA is generally better able to cope with such variation compared with more traditional immunoassays, because although epitopes might alter, requiring different antibodies for successful detection, the DNA/RNA tends to be more conserved and thus several variants that would require different antibodies for detection may be detectable using the same pair of RPA primers.

The central processing unit may control the processing of samples within the reaction vessels, preferably controlling the heating of the samples. Typically, the central processing unit controls the processing of one or more reaction vessels independently and/or concurrently.

To achieve a high throughput of screening it is preferred that an individual reaction vessel is processed to yield a result in less than about 60 minutes, preferably in less
than about 15 minutes, less than about 12 minutes, less than about 10 minutes, less than about 9 minutes, less than about 8 minutes, less than about 7 minutes, less than about 6 minutes, less than about 5 minutes.

Preferably, an individual reaction vessel is processed to yield a result in at least about 1 minute, at least about 2.5 minutes, at least about 5 minutes, at least about 7 minutes, at least about 8 minutes, at least about 9 minutes, at least about 10 minutes, at least about 12 minutes, at least about 15 minutes, at least about 20 minutes, at least about 30 minutes, at least about 40 minutes, at least about 50 minutes, at least about 60 minutes.

In embodiments, an individual reaction vessel is processed to yield a result in from about 1 minute to about 60 minutes, 5 minutes to about 30 minutes, preferably from about 7 minutes to about 15 minutes, more preferably from about 8 minutes to about 12 minutes. In embodiments, an individual reaction vessel is processed to yield a result in from preferably from about 1 minute to about 10 minutes, from about 1 minute to about 8 minutes, from about 1 minute to about 5 minutes, from about 1 minute to about 2.5 minutes, from about 2.5 minutes to about 5 minutes, from about 2.5 to about 8 minutes, from about 2.5 to about 10 minutes.

Different reaction vessels may require different processing times depending on their target, their specificity, the reagents, the sample quality, and such forth. Having identified the reaction vessel, the central processing unit will typically select a predetermined set of conditions appropriate for the identified reaction vessel, e.g. heating to the sample and reagents within a reaction chamber of the reaction vessel to a predetermined temperature.

Reaction vessels suitable for use in the invention will typically comprise one, two, three, four, five or more reaction chambers. Each reaction chamber will usually perform a different assay relating to a different pathogen. Usually, the same sample will be tested in each reaction chamber of a reaction vessel. The reaction chamber will typically contain reagents for performing the assay.
As discussed in other aspects of the invention, which discussion is equally applicable to this aspect, a reaction vessel may be identified by information presented on the reaction vessel, preferably wherein the information is presented in a bar code, a smart chip, an RFID tag or a key. The bar code may be a linear bar code, a 2D bar code, a QR code, an alpha-numeric code.

The information may be acquired by direct contact between the identification device and the information presented on the reaction vessel. Typically, the coded information is acquired by indirect contact between the identification device and the information presented on the reaction vessel.

The identification device may comprise a coded information reader in communication with the central processing unit. The coded information reader may be integrally formed within the control module or the process modules. Alternatively, the coded information reader may be a separate module.

The sample may be a biological sample, a patient sample, a veterinary sample, or an environmental sample. The sample may be raw (e.g. a crude matrix) or optionally processed.

Typical samples include those containing spores, viruses, cells, nucleic acid from prokaryotes or eukaryotes, or any free nucleic acid. The sample(s) may be isolated from any material suspected of containing a target pathogen.

For example, for animals, for example, mammals, such as, for example, humans, the sample may comprise blood, bone marrow, mucus, lymph, hard tissues, for example, liver, spleen, kidney, lung, or ovary, biopsies, sputum, saliva, tears, faeces, or urine. Or, the sample may be taken from air, plant, soil, or other materials suspected of containing biological organisms.

Samples may also contain environmental and contaminants such as dust, pollen, and soot (for example, from diesel exhaust), or clinically relevant matrices such as urine, mucus, or saliva.
Samples may also include waste water, drinking water, air, milk, or other food.

Depending on the concentration of contaminants, sample purification methods known to those of ordinary skill in the art may be required to remove inhibitors for the assays. Purification may, for example, involve the use of detergent lysates, sonication, vortexing with glass beads, or a French press. This purification could also result in concentration of the sample. Samples may also be further purified, for example, by filtration, phenol extraction, chromatography, ion exchange, gel electrophoresis, or density dependent centrifugation.

In particular embodiments, the sample can be added directly to the reaction reagents, or pre-diluted and then added to the reaction mix, in either case without prior purification of target nucleic acid.

The invention also contemplates a reaction vessel located in the modular system and/or a modular system comprising at least one first reaction vessel and the at least one second reaction vessel and optionally at least one third reaction vessel.

The invention further provides a kit for performing concurrent assays on a sample, the kit comprising a modular system according to previously discussed aspects of the invention.

The kit may comprise a plurality of first reaction vessels, each configured to perform a predetermined specific assay or assays on a sample and suitable for use in the at least one first process module; a plurality of second reaction vessels, each configured to perform a predetermined specific assay or assays on a sample and suitable for use in the at least one second process module; and optionally a plurality of further reaction vessels, each configured to perform a predetermined assay or assays on a sample and suitable for use in at least one of the optional further process modules.

The first, second and optional third reaction vessels may each be configured to identify a different target analyte or group of target analytes, typically a different pathogen or group of pathogens.
The invention further provides a method of using the modular system for performing concurrent assays on at least one sample to investigate the content of said at least one sample.

A method for identifying a pathogen or other target analyte in a sample using a plurality of different assays may comprise the steps of: collecting, and optionally preparing, a sample to be tested; selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an assay; selecting process modules for receiving and processing the selected reaction vessels, each process module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel; gathering information relating to the identity of each reaction vessel and/or each sample; functionally adjoining the process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or other target analytes in the sample; loading each reaction chamber with the sample; providing conditions suitable for performing the assays; gathering information relating to the assay output in the reaction chambers; and identifying pathogens (or other target analytes) present in the sample by processing the information gathered using the central processing unit.

Typically, each separate reaction vessel identifies a different pathogen (or other target analyte) or different group of pathogens (or different group of target analytes).

Additionally, or alternatively, the plurality of reaction vessels is selected from a larger group of reaction vessels, wherein each reaction vessel is configured to perform at least one assay to identify at least one target pathogen pathogen (or other target analyte) or group of pathogens (or group of target analytes). Typically, the group of reaction vessels is capable of identifying a plurality of different pathogens (or other target analytes) or groups of pathogens (or other target analytes). Typically, the group of reaction vessels is capable of identifying a greater number of pathogens (or other target analytes) or groups of pathogens (or other target analytes) than the number that can be identified by the selection of reaction vessels. The user may select from the
group of reaction vessels depending upon the identity of the pathogen(s) (or other target analytes) of interest and/or identity of the pathogen(s) (or other target analytes) suspected of being in the sample.

The assays may be performed independently and/or concurrently.

Additionally, or alternatively, the invention provides a method for identifying the prevalence of a target pathogen, or pathogens, or other target analytes, in a selection of different samples using a plurality of assays.

The method may comprise the steps of: collecting, and optionally preparing, a plurality of different samples to be tested; selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an assay; selecting process modules for receiving and processing the selected reaction vessels, each process module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel; gathering information relating to the identity of each reaction vessel and/or each sample; functionally adjoining the process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or other target analytes in the sample; loading each reaction chamber with a different sample; providing conditions suitable for performing the assays; gathering information relating to the assay output in the reaction chambers; identifying which of the samples contain the pathogen or other target analytes by processing the information gathered using the central processing unit.

Typically, each of the reaction vessels is configured to identify the same pathogen or group of pathogens (or other target analytes). Said reaction vessels may be selected from a larger group of reaction vessels wherein at least a portion of reaction vessels making up said group are configured to perform assays for identifying different target pathogens (or other target analytes).

The assays may be performed independently and/or concurrently.
The modular systems for performing concurrent assays according to the invention may be used in the previously describe methods.

5 Brief Description of the Figures

The present invention may be carried out in various ways and preferred embodiments of systems, devices, kits and methods will now be described with reference to the accompanying drawings in which:

10 Fig. 1 shows a system according to the invention.

Fig. 2 shows a reaction vessel, sample reservoir and liquid transfer device.

15 Fig. 3 shows an alternative reaction vessel, sample reservoir and liquid transfer device.

Fig. 4 shows a system according to the invention.

20 Fig. 5 shows a system according to the invention.

Fig. 6 shows a system according to the invention.

Fig. 7 shows a system according to the invention.

25 Fig. 8 shows a system according to the invention.

Fig. 9 shows a system interface.

30 Fig. 10 shows a portion of a system interface.

Fig. 11 shows an alternative system interface.
Fig. 12 shows a modular system for performing concurrent isothermal nucleic acid amplifications.

Fig. 13 shows a flow diagram of a method according to the invention.

Fig. 14 shows a flow diagram of an alternative method according to the invention.

Fig. 15 to 20 show modular systems according to the invention.

Fig. 21 shows a flow diagram of a method according to the invention.

Fig. 22 shows a flow diagram of an alternative method according to the invention.

**Detailed Description**

The present invention provides systems, devices, kits and methods for performing point of care isothermal nucleic acid amplification assays, in particular for performing a plurality of concurrent isothermal nucleic acid amplification assays in a point of care environment.

By "isothermal" is meant a set of reaction conditions where the temperature of the reaction is kept essentially or substantially constant during the course of the amplification reaction. An advantage of the amplification method of the present methods is that the temperature does not need to be cycled between an upper temperature and a lower temperature. The reactions used in the invention will work at the same temperature or within the same narrow temperature range. However, it is not necessary that the temperature be maintained at precisely one temperature. If the equipment used to maintain an elevated temperature allows the temperature of the reaction mixture to vary by a few degrees, or few tenths of a degree, such as, for example, less than 1 degree, 0.8 degrees, 0.6 degrees, 0.4 degrees, or 0.2 degrees, this is not detrimental to the amplification reaction, and may still be considered to be an isothermal reaction.
The systems and methods disclosed herein may be performed on or use a crude matrix. As used herein, a "crude matrix" is a matrix that includes nucleic acids from a biological source, wherein the matrix has not been subject to nucleic acid extraction and/or purification. In some embodiments, the biological source includes cells and or biological sample (e.g. from a patient) and/or an environmental sample.

The crude matrix may include a biological sample, e.g., a sample obtained from a plant or animal subject. As used herein, biological samples include all clinical samples useful for detection of nucleic acids in subjects, including, but not limited to, cells, tissues (for example, lung, liver and kidney), bone marrow aspirates, bodily fluids (for example, blood, derivatives and fractions of blood (such as serum or buffy coat), urine, lymph, tears, prostate fluid, cerebrospinal fluid, tracheal aspirates, sputum, pus, nasopharyngeal aspirates, oropharyngeal aspirates, saliva), eye swabs, cervical swabs, vaginal swabs, rectal swabs, stool, and stool suspensions. Other suitable samples include samples obtained from middle ear fluids, bronchoalveolar lavage, tracheal aspirates, sputum, nasopharyngeal aspirates, oropharyngeal aspirates, or saliva. In particular embodiments, the biological sample is obtained from an animal subject. Standard techniques for acquisition of such samples are available. See for example, Schluger et al, J. Exp. Med. 176: 1327-33 (1992); Bigby et al, Am. Rev. Respir. Dis. 133:515-18 (1986); Kovacs et al, NEJM 318:589-93 (1988); and Ognibene et al, Am. Rev. Respir. Dis. 129:929-32 (1984). In some embodiments, the crude matrix includes an environmental sample, e.g., a surface sample (e.g., obtained by swabbing or vacuuming), an air sample, or a water sample.

The crude matrix may include isolated cells, e.g., animal, bacterial, fungal (e.g., yeast), or plant cells, and/or viruses. The isolated cells can be cultured using conventional methods and conditions appropriate for the type of cell cultured.

WO2011/038197 discloses methods for performing isothermal nucleic acid amplifications on crude matrix samples and is incorporated herein by reference.

Fig. 1 shows a system (1) according to the invention. The system comprises a plurality of ports (2) each configured to receive a reaction vessel (not shown). The reaction vessels are configured to perform an isothermal nucleic acid amplification assay on a
sample when received in a port (2). In this embodiment, the reaction vessels are in the form of cassettes which are received by the ports (2). The illustrated system (1) has twenty-four ports arranged in three banks of eight. The system can independently process up to 24 cassettes concurrently.

The system also includes means for identifying each reaction vessel and means for identifying the sample to be tested located in the reaction vessel. In the system of this embodiment, identification code readers are located within the ports: one per port (2) for identifying the reaction vessels once they are located in the ports (2). A separate identification code reader, similar to that shown in Fig. 5 (148) is used to identify patient samples. Each port (2) also includes an individually controlled heating element.

The system also includes detectors (not shown) for gathering information relating to amplified nucleic acid in each reaction vessel. Again, the detectors are located within the ports (2): typically, one detector per port (2). The system also includes a built-in computer (3) containing central processing unit (not shown) programmed to process the information gathered relating to amplified nucleic acid in each identified reaction vessel. The built-in computer (3) also includes user interface (4) for presenting information related to the amplified nucleic acid from each sample. In this instance, the user interface is a touch screen monitor (4). The system also includes a number of data ports (5) and a power supply port (6).

In use, the operator will select cassettes suitable for performing isothermal nucleic acid amplifications to identify a pathogen or pathogens of interest. Each cassette is loaded with a sample and the identity of the sample in each individual cassette is identified and recorded using a barcode reader. The cassettes are then placed into the ports, where a second coded information reader identifies the cassettes. The central processing unit controls the conditions in each reaction vessel according to the cassette’s specific identity in order to attempt to perform an isothermal nucleic acid amplification on the sample contained within the cassette. Whilst the samples are processed detectors gather data relating to amplified nucleic acid inside each cassette. The data gathered is processed in the built-in computer and results displayed on the built-in monitor. The cassettes self-seal at the end of the process so that they can be disposed of safely.
In the exemplified embodiment, up to twenty-four cassettes can be processed concurrently, although not all the ports need to be in use at any one time. The invention is not limited to twenty-four ports, systems with more or less ports are also contemplated. The illustrated system can process up to twenty-four cassettes all containing the same sample concurrently. For instance, each cassette can be different (e.g. test for a different pathogen(s)/target sequences) and all contain the same sample. Alternatively, a combination of samples and cassettes can be processed concurrently. The sample is loaded into the cassette from a sample reservoir using a liquid transport device. Typically, each cassette will have from 1 to 4 reaction chambers for processing the sample. Typically, each cassette can perform from 1 to 4 isothermal nucleic acid amplifications, preferably in order to test for from 1 to 4 different bacterial or viral targets.

The system may be used with cassettes configured for NEAR ( nicking and extension amplification reaction) and/or RPA (recombinase polymerase amplification) isothermal nucleic acid amplifications. Accordingly, the cassettes’ reaction chambers may contain reagents necessary for performing NEAR and/or RPA on samples introduced into said chambers. Typically, the reagents will be dry.

Nicking and extension amplification reactions are discussed in detail in WO2009/012246 which is incorporated herein by reference.


The system can process NEAR and/or RPA isothermal nucleic acid amplifications concurrently.

Fig. 4 shows an alternative system according to the invention. Fig. 2 shows a reaction vessel (200), sample reservoir (300) and liquid transfer device (100) suitable for use in the system illustrated in Fig. 4. Each subassembly can have a D-shaped or
otherwise asymmetrical cross section (105, 205, 305) that is compatible with the other two subassemblies, such that the subassemblies may only be mated to each other in one orientation.

The reaction chamber 200 includes a microtube 220 held within an aperture in the bottom of the reaction vessel body 210.

Fig. 2 shows the transfer device 100 and reaction vessel 200 as described above with one pipette tip 120 and one microtube 220. FIG. 3 shows the transfer device 100 and amplification chamber 200 with two pipette tips 120 and two microtubes 220. Using the device in FIG. 3, different amplification reactions can be performed on two portions of one sample.

Such assemblies are discussed in detail in WO2013/041713 which is incorporated herein by reference.

Fig. 4 shows an alternative system (40) according to the invention. The illustrated system comprises a series of four sample processing devices (42a, 42b, 42c, 42d). The devices each contain a port (not shown) for receiving the reaction vessels illustrated in Fig. 3 and Fig. 5. Each device also contains a detector for gathering data relating to amplified nucleic acid and barcode reader for identifying the reaction vessels by means of a barcode located thereon. Each device (42a, 42b, 42c, 42d) has its own independent power supply (50b, 50c, 50d, 50e) and is in wired communication with a network hub (52). The network hub is, in turn, in wired communication with a computer (44) containing a central processing unit which is programed to process the data gathered by the detectors and present the results on its monitor. The computer (44) is also in wired communication with a barcode reader (48) for identifying the samples being tested and a printer (46) for printing the results. The computer (44) has its own independent power source (50a) although it may equally run off a battery.

Suitable sample processing devices are available from Alere Inc. under the brand name Alere i. Suitable computers are widely available.
The system may be used with reaction vessels configured to perform NEAR and/or RPA isothermal nucleic acid amplifications. Accordingly, the cassettes' reaction chambers may contain reagents necessary for performing NEAR and/or RPA on samples introduced into said chambers.

The system can process NEAR and/or RPA isothermal nucleic acid amplifications concurrently, either in separate reaction vessels or within separate reaction chambers of a single reaction chamber or both.

Fig. 5 shows an alternative system (140) according to the invention. The illustrated system comprises a series of four sample processing devices (142a, 142b, 142c, 142d). This system differs from that shown in Fig. 4 in that the sample processing devices (142a, 142b, 142c, 142d) are docked with a docking station (150c). The docking station has its own independent power supply (150a) and supplies power to the sample processing devices (142a, 142b, 142c, 142d). Preferably, the sample processing devices (142a, 142b, 142c, 142d) are separated so as to allow access to their power buttons. In this embodiment, the docking station (150c) is in wired communication with a computer (144) preprogramed to process the data gathered by the sample processing devices (142a, 142b, 142c, 142d), a barcode reader (148) and a printer (146). The computer (144) has its own independent power supply (150b). This system has the advantage of being easy to assemble.

Fig. 6 shows a still further embodiment of the system of the invention. In this instance, the system (240) comprises a docking station (250c) and four sample processing devices (142a, 142b, 142c, 142d) with which it is in communication. The docking station has its own power supply (250a) and is in wired communication with a barcode reader (248) and a printer (246). The docking station also includes a dock for receiving a tablet computer (244). The tablet computer (244) has a central processing unit and is preprogramed for processing the data gathered by the sample preparation devices. The tablet computer (244) is in wireless communication with the docking station and, thereby, the four sample processing devices, the printer and the barcode reader. When received in the tablet dock on the docking station (250c), the tablet computer (244) may be recharged by the docking station’s power supply.
Fig. 7 shows a still further embodiment of the invention. In this embodiment, the system (340) comprises a single sample processing device (242a). The device has four ports for receiving a reaction vessel (342a, 342b, 342c, 342d). The ports are identical in nature to those describe above in relation to Fig. 4. The device is in wired communication with a computer (344). The computer (344) and device (242a) have their own independent power supplies (350a and 350b respectively).

Fig. 8, 10 and 11 show alternative system interfaces. They are particularly suitable for use in the embodiment illustrated in Fig. 4.

In the arrangement illustrated in Fig. 8 and 9, a central hub (452) is used to provide power and data connectivity to the sample processing devices (not shown). The hub is in wired communication with a computer (444) and has its own dedicated power supply (450). The computer (444) is in wired communication with a barcode scanner (448). Power and data connectivity are provided to the sample processing devices by means of the interconnecting dual power/data supply cables 460a, 460b, 460c and 460d, which form a backbone connector (460). Each dual cable includes a dual power/data input cable (461) and a dual power/data output cable (462), and a power output cable (464) and a data output cable (463). The power output cable (464) and data output cable (463) terminate with connectors (466, 465) suitable for connecting to the sample processing devices; thereby providing power and data connectivity to the sample processing devices so connected. The dual power/data input cable (461) terminates with a connector (467) suitable for connecting to the hub and/or the connector (468) which terminates the dual power/data output cable (462).

As shown, each dual input cable is capable of being received by and connecting with a dual output cable. This enables multiple interconnecting dual cables to be joined in series (460) and thereby link multiple sample processing devices to the computer (444). Adding an extra sample processing module to the series requires only an additional dual interconnecting cable (460c) to be attached to the last dual interconnecting cable in the series (460d). This system interface enables the system in Fig. 4. to be expanded and contracted in a very straightforward manner.
Fig. 10 illustrates an alternative arrangement to that shown in Fig. 8. In this embodiment, the hub and computer have been replaced by a dedicated computer terminal with a built-in display (544). The dedicated computer terminal (544) has an independent power supply (550) and is connected to a barcode reader (548). The system includes a backbone (560) of four interconnecting dual power/data cables (560a, 560b, 560c, 560d). Depending on the exact configuration of the system, the dedicated computer terminal (544) may control sample processing devices connected thereto and/or process data gathered therefrom. The dedicated computer terminal (544) provides power to any sample processing devices connected to it by means of the interconnecting dual power/data cables (560a, 560b, 560c, 560d).

Fig. 11 shows an alternative arrangement. In this arrangement the dual data/power cables (660a, 660b, 660c and 660d) are not interconnected. Each dual data/power cable has a dual input cable, but separate power and data output cables for connecting to the sample processing devices (not shown). Each dual data/power cable is connected to a hub (652) which in turn has a dedicated power supply and is in communication with a computer (644). The computer (644) is in communication with a barcode reader (648). Depending on the embodiment of the invention, the computer (644) may control sample processing devices connected thereto and/or process data gathered therefrom.

Fig. 12 shows a modular device (20) according to the invention. The device provides high throughput screening using isothermal nucleic acid amplification for the determination of microbial and viral content of a sample, typically at or close to the point of sampling. The device (20) comprises a control module (22); a plurality of process modules (24, 26) in operable communication with the control module (22); a user interface (27); a communication port (not shown) and a data display (28). Each process module can hold a reaction vessel as shown in Fig. 2 or Fig. 3, and includes a heating element and a detector for gathering data relating to amplified nucleic acid. The process module may be configured to perform recombinase polymerase amplification of nucleic acid and/or to perform nicking and extension amplification reaction to amplify nucleic acid.
This modular device is particularly advantageous because there is no limit to the number of process modules (24, 26) that can be connected to the control module (22). The connection between each module (22, 24, 26) provides both power from the control module (22) and data connectivity with the control module (22).

Fig. 13 and 14 set out alternative processes according to the invention.

Fig. 13 shows a method for identifying a target sequence, typically from a target pathogen, in a single sample using isothermal nucleic acid amplification. This method may be implemented using any of the kits and/or systems of the invention.

The first step is collecting and, optionally, preparing a sample to be tested. The sample is usually a biological sample, a patient sample, a veterinary sample, or an environmental sample. Typically, the optional sample preparation includes mixing the sample with an elution buffer in a sample reservoir.

Once the sample has been collected and optionally prepared, the user selects a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an isothermal nucleic acid amplification. For the purpose of the invention, it is understood that “separate reaction vessels” means that the reaction vessels are physically separate (i.e. they do not for part of a single structure or unitary device). The reaction vessels are usually chosen based upon the target viruses and bacteria which are under investigation. In embodiments of the invention, the selection will be made based upon disease symptoms displayed by a patient. By matching the reaction vessels to symptoms and processing samples concurrently, the user can make an accurate diagnosis very quickly.

The prepared samples are loaded into the selected reaction chambers of the selected reaction vessels. The reaction chambers will typically contain dry reagents for performing isothermal nucleic acid amplifications, typically NEAR or RPA reaction. Typically, each reaction vessel identifies a different pathogen/target sequence or different group of pathogens/target sequences.
Samples and reagents are subsequently exposed to conditions suitable for performing isothermal nucleic acid amplification concurrently. Typically, this will involve heating the contents of reaction chambers (e.g. sample, buffers and reagents) to their isothermal set point concurrently. The isothermal set point is the temperature at which the isothermal amplification is performed. Different reaction chambers will require heating to different temperatures depending upon the amplification they are intended to perform. Typically, all of the reaction chambers of a single reaction vessel will require heating to the same temperature. The reaction chambers will be heated for sufficient time for the isothermal amplification to occur, progress may be monitored using an appropriate detector. The amount of time required will depend on the sample and the specific amplification being performed.

Once the samples and reagents have been exposed to conditions suitable for performing isothermal nucleic acid amplification, information is gathered relating to amplified nucleic acid in the reaction chambers. This will usually be performed using an appropriate detector, such as a fluorescence detector. The information may be gathered continually throughout the heating of the sample, after a period of time sufficient for amplification to occurred, or both during and after the time amplification may have occurred.

The data gathered will be processed using a preprogrammed central processing unit in order identify any viruses and/or bacteria present.

Fig. 14 is a flow diagram illustrating a method for identifying the prevalence of target sequences or pathogens in a selection of different samples using isothermal nucleic acid amplification. Again, this method may be implemented using any of the kits and/or systems of the invention.

The method involves first collecting a plurality of different samples to be tested. The samples may or may not require preparation before mixing with the amplification reagents, typically this will involve mixing the sample with an elution buffer in a sample reservoir. The sample is usually a biological sample, a patient sample, a veterinary sample, or an environmental sample.
The user will then select a plurality of separate reaction vessels from a larger group of separate reaction vessels, each reaction vessel being configured to perform at least one isothermal nucleic acid amplification in a reaction chamber on a target sequence. In a preferred implementation of the method, each of the reaction vessels which are selected are capable of performing the same isothermal nucleic acid amplification(s).

Each reaction vessel is then loaded with a different sample. Reaction chambers within a single reaction vessel may be loaded with the same sample.

In steps similar to those described in relation to Fig. 13, the user then provides conditions suitable for performing isothermal nucleic acid amplifications in each reaction chamber, preferably concurrently, and gathers information relating to amplified nucleic acid in the reaction vessels.

The information gathered is then processed using a preprogrammed central processing unit, which identifies which of the samples contain the target viruses and/or bacteria.

It will be appreciated that investigations using the methods disclosed in Fig. 13 and Fig. 14 may themselves be practiced concurrently on a single system according to the invention.

Accordingly, the present invention provides systems, devices, kits and methods for performing point of care isothermal nucleic acid amplifications, in particular for performing a plurality of concurrent isothermal nucleic acid amplifications in a point of care environment.

The present invention also provides systems, devices, kits and methods for performing point of care assays, in particular for performing concurrent assays on at least one sample to investigate the biological content thereof in a point of care environment.

Fig. 15 a-c show an assembled modular system, and a control module and two process modules.
Fig. 15a shows a first process module (151) and a second process module (152). The first (151) and second (152) process modules are different. The first process (151) module is configured to receive up to two reaction vessels in two ports (153, 154), whereas the second process module (152) is configured to receive a single reaction vessel in a single port (155).

Fig. 15b shows a control module (156) for receiving the process modules (151, 152) in Fig. 15b. The control module comprises a housing (157) configured to receive at the process modules (151, 152) within said housing (157), the central processing unit (not shown), a display interface (158) in a folded down storage arrangement. The process modules (151, 152) are detachably and slidably receivable within the housing (157). The housing (157) includes a series of horizontal runners (159) on an inner wall thereof for receiving the process modules (151, 152). The runners (159) are in the form of elongate protrusions and are substantially equally spaced along two inner walls of the housing (157).

Fig. 15c shows an assembled system according to the invention comprising a first (1510), second (1511) and third (1512) process modules, each received within the housing (1513) of a control module (1514) comprising a display interface (1515). The three process modules (1510, 1511, 1512) are different and each is configured to perform a different assay(s). The first process module (1510) comprises two ports (1516, 1517) for receiving up to two reactions vessels concurrently, whereas the second and third process modules (1511, 1512) each comprise one port (1518, 1519) for receiving a single reaction vessel. In the system illustrated in Fig. 15c at least four assays can be performed independently and/or concurrently. The display interface (1515) is a touchscreen display and is used for inputting control commands to the system and to display the output of the assays.

Fig. 16 shows an alternative modular system according to the invention (160). The system (160) comprises a control module (161) comprising the central processing unit, the display interface (162) and identification device (163). In this system, the first (164) and second (not shown) process modules and the control module (161) detachably connect to one another. The first process module (164) comprises a port (166) configured to receive a reaction vessel (not shown).
The modular system further comprises a power module (165) for powering the process modules and the control module. The illustrated power module comprises a battery (167). The power module (165) detachably connects to any one of the process modules to provide power thereto.

The process module (164), control module (161) and power modules (165) are detachably connectable in a vertical stack arrangement, with the control module (161) located at a top of the stack, and the power module (165) located at a base of the stack. The control module comprises four male physical connection means on its base, of which three are visible (168, 169, 1610). The power module (165) comprises four female physical connection means on its upper surface, of which three are visible (1611, 1612, 1613). The process module comprises physical connection means both on its upper surface (1617, 1618, 1619) and its base (1614, 1615, 1616) for receiving the physical connection means of the power module and the control module. The physical connection means include electronic connection means for provide power to the control and process modules. The process modules are in wireless communication with the control module.

Fig. 17 shows an alternative modular system according to the invention (170). In this system (170) the process modules (171, 172) and in wired communication with the control module (174) by means of a ribbon cable (175). The ribbon cable (175) also provides power to the control module (174) and the process modules (171, 172) from the power module (173). In this system, the power module (173) is mains powered (176). The power module (173) also include cable sockets (177, 178) so that the system (170) may be connected to similar modular systems or to a computer network.

Fig. 18 shows stackable process modules (181, 182) for use in all aspects of the invention. Each process module (181, 182) includes a port (183, 184) for receiving a reaction vessel (not shown) and a display interface (185, 186). Also provided is a storage module (187) for storing equipment useful in the performance of assays and/or the preparation of samples. The storage module (187) also supports the process module (181) located above, allowing the process modules (181, 182) to be stacked in the illustrated horizontally offset manner.
Fig. 19 shows alternative stackable process modules (191, 192) for use in all aspects of the invention. Each process module (191, 192) includes a port (193, 194) for receiving reaction vessels (195, 196), such as reaction cassettes; a display interface (197, 198); and a storage compartment or drawer (199, 1910) for storing equipment (1911) useful in the performance of assays and/or the preparation of samples. Each process module (191, 192) also includes means for identifying the sample or reaction vessel, such as a barcode reader (1912, 1913).

Fig. 20 shows storage modules (201, 202) for use in conjunction with process modules (203, 204) of the invention. The storage modules (201, 202) allow for convenient storage of equipment useful in the performance of assays and/or the preparation of samples. Typically, the storage modules (201, 202) comprise a hollow body with a closable door (205, 206). Preferably, the storage modules (201, 202) have substantially the same outer shape as the process modules. Preferably the storage modules (201, 202) and process modules (203, 204) are stackable. The user may therefore build an array of process and storage modules depending on their needs.

Fig. 21 and 22 set out further processes according to the invention.

Fig. 21 shows a method for identifying a target biological marker, typically from a target pathogen, or other target analyte in a single sample using plurality of different assays. This method may be implemented using any of the kits and/or systems of the invention.

The first step is collecting and, optionally, preparing a sample to be tested. The sample is usually a biological sample, a patient sample, a veterinary sample, or an environmental sample. Typically, the optional sample preparation includes mixing the sample with an elution buffer in a sample reservoir.

Once the sample has been collected and optionally prepared, the user selects a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an assay. For the purpose of the invention, it is understood that “separate reaction vessels” means that the reaction vessels are physically separate (i.e. they do not for part of a single structure or unitary
device). The reaction vessels are usually chosen based upon the target viruses and/or bacteria and/or other target analytes which are under investigation. In embodiments of the invention, the selection will be made based upon disease symptoms displayed by a patient. By matching the reaction vessels to symptoms and processing samples concurrently, the user can make an accurate diagnosis very quickly.

The user will then select process modules for receiving and processing the selected reaction vessels, each process module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel.

Next, the user will functionally adjoin the selected process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or other target analytes in the sample.

The samples are loaded into the selected reaction chambers of the selected reaction vessels. The reaction chambers will typically contain dry reagents for performing the assays, for instance a NEAR or RPA reaction. Typically, each reaction vessel identifies a different pathogen/target sequence or different group of pathogens/target sequences or other target analytes.

Samples and reagents are subsequently exposed to conditions suitable for performing the assays concurrently. Typically, this may involve heating the contents of reaction chambers (e.g. sample, buffers and reagents) concurrently to the temperature at which the assay is performed. Different reaction chambers will require heating to different temperatures depending upon the assay they are intended to perform. Some may require no heating at all. Typically, all of the reaction chambers of a single reaction vessel will require heating to the same temperature. The reaction chambers will be heated for sufficient time for the assay to occur, progress may be monitored using an appropriate detector. The amount of time required will depend on the sample and the specific assay being performed.
Once the samples and reagents have been exposed to conditions suitable for performing the assay, information is gathered relating to markers in the reaction chambers. This will usually be performed using an appropriate detector, such as a fluorescence detector. The information may be gathered continually throughout the performance of the assay, after a period of time sufficient for the assay reaction to have occurred, or both during and after the time the assay reaction occurred.

The data gathered will be processed using a preprogrammed central processing unit in the control module in order to identify any viruses and/or bacteria and/or other target analytes present.

Fig. 22 is a flow diagram illustrating a method for identifying the prevalence of target biological marker or pathogen(s) or other target analyte(s) in a selection of different samples using a plurality of assays. Again, this method may be implemented using any of the kits and/or systems of the invention.

The method involves first collecting a plurality of different samples to be tested. The samples may or may not require preparation before mixing with the assay reagents, typically this will involve mixing the sample with an elution buffer in a sample reservoir. The sample is usually a biological sample, a patient sample, a veterinary sample, or an environmental sample.

The user will then select a plurality of separate reaction vessels from a larger group of separate reaction vessels, each reaction vessel being configured to perform at least one assay in a reaction chamber on a target biological marker/analyte. In a preferred implementation of the method, each of the reaction vessels which are selected are capable of performing the same assay(s).

Each reaction vessel is then loaded with a different sample. Reaction chambers within a single reaction vessel may be loaded with the same sample.

In steps similar to those described in relation to Fig. 21, the user then selects process modules for receiving and processing the selected reaction vessels, each process
module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel.

Next, the user will functionally adjoin the selected process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or other target analytes in the sample.

The process modules then provide conditions suitable for performing the assays in each reaction chamber, preferably concurrently, and gather information relating to biological markers and/or other target analytes in the reaction vessels.

The information gathered is then processed using a preprogramed central processing unit, which identifies which of the samples contain the target viruses and/or bacteria and/or other target analytes.

It will be appreciated that investigations using the methods disclosed in Fig. 21 and Fig. 22 may themselves be practiced concurrently on a single system according to the invention.

It will be appreciated that various modifications may be made to the embodiments shown without departing from the spirit and scope of the invention as defined by the accompanying claims as interpreted under patent law.
What is claimed:

1. A system for performing concurrent isothermal nucleic acid amplification assays, the system comprising:
   a. a plurality of ports each configured to receive a reaction vessel, each reaction vessel being configured to perform an isothermal nucleic acid amplification assay on a target sequence in a sample when received in said port;
   b. at least one identification device for gathering information relating to the identity of the reaction vessel and/or the sample;
   c. at least one detector for gathering information relating to amplified nucleic acid in each reaction vessel;
   d. a central processing unit programmed to process the information gathered relating to amplified nucleic acid in each said reaction vessel and the information gathered relating to the identity of the sample and/or reaction vessel; and
   e. a display interface in communication with the central processing unit which presents information relating to the amplification of nucleic acid in each sample.

2. The system according to claim 1 wherein the system is configured to receive and process a plurality of reaction vessels independently.

3. The system according to claim 1 or 2 wherein the central processing unit controls the processing of samples within the reaction vessels, preferably the heating of the samples, preferably the independent heating of reaction vessels.

4. The system of claim 1 to 3 wherein the central processing unit controls the processing of one or more reaction vessels independently and concurrently to yield rapid analysis of samples containing target nucleic acid sequences for amplification.
5. The system of any one of claims 1 to 4 wherein an individual reaction vessel is processed to yield a result in less than 15 minutes, less than 12 minutes, less than 10 minutes, less than 9 minutes, less than 8 minutes, less than seven minutes, less than six minutes, less than five minutes.

6. The system of claim 5 wherein an individual reaction vessel is processed to yield a result in at least about 5 minutes, at least about 7 minutes, at least about 8 minutes, at least about 9 minutes, at least about 10 minutes, at least about 12 minutes, at least about 15 minutes, at least about 20 minutes, at least about 30 minutes.

7. The system of any one of claims 1 to 6 wherein a reaction vessel is identified by information presented on the reaction vessel, preferably wherein the information is presented in a bar code, a smart chip, an RFID tag or a key.

8. The system of claim 7 wherein the bar code is a linear bar code, a 2D bar code, a QR code, an alpha-numeric code.

9. The system of claim 7 wherein the information is acquired by direct contact between the identification device and the information presented on the reaction vessel.

10. The system of claim 7 wherein the coded information is acquired by indirect contact between the identification device and the information presented on the reaction vessel.

11. The system of any one of claims 1 to 10 wherein the identification device comprises a coded information reader in communication with the central processing unit.

12. The system of any one of claims 1 to 11 wherein the sample is a biological sample, a patient sample, a veterinary sample, or an environmental sample.
13. The system of any one of claims 1 to 12 comprising a wired and/or wireless connection for transmitting the information relating amplified nucleic acid from the detector to the central processing unit.

14. The system according to any preceding claim wherein the reaction vessels comprise one or more sample chambers for containing the sample or samples to be processed.

15. The system of claim 1 comprising a plurality of devices each comprising at least one port and wherein each device is connected wirelessly or wired or located on a docking station.

16. A reaction vessel received in a system according to any preceding claim.

17. The reaction vessel according to claim 16 comprising the reagents for a recombinase polymerase amplification (RPA).

18. The reaction vessel according to claim 16 comprising the reagents for a nicking and extension amplification reaction (NEAR).

19. The reaction vessel of claim 17 wherein the RPA reagents comprise a recombinase, a single strand binding protein and a polymerase.

20. The reaction vessel of claim 19 wherein the recombinase is selected from T4 UvsX, T6 UvsX, or RecA.

21. The reaction vessel of claim 19 or 20 wherein the polymerase is selected from the group consisting of E. coli DNA polymerase I Klenow fragment, B. stearothermophilus polymerase (Bst), B. subtilis Phi-29 polymerase, B. subtilis polymerase I (Bsu).

22. The reaction vessel of claim 19, 20 or 21 wherein the single strand binding protein is gp32.
23. The reaction vessel of any one of claims 19 to 22 further comprising a crowding agent, ATP or an ATP analog, dNTP(s), or T4 bacteriophage UvsY.

24. The reaction vessel of claim 23 wherein the crowding agent is selected from the group consisting of polyethylene glycol (PEG), dextran, polyvinylalcohol (PVA), polyvinylpyrrolidone (PVP) or Ficoll.

25. The reaction vessel of claim 24 wherein the PEG is PEG1450, PEG3000, PEG8000 or PEG10000.

26. The reaction vessel of claim 24 wherein the PEG has a molecular weight between 15000 and 20000.

27. The reaction vessel according to any one of claims 23 to 26 wherein the dNTP(s) is/are selected from the group consisting of dATP, dGTP, dCTP, dTTP.

28. The reaction vessel any one of claims 23 to 27 wherein the ATP or ATP analog is selected from ATP, ATP-γ-S, ATB-β-S, ddATP or a combination thereof.

29. The reaction vessel of claim 18 wherein the NEAR reagents comprise a nicking enzyme, a forward template nucleic acid, a reverse template nucleic acid, and a polymerase.

30. The reaction vessel according to any one of claims 16 to 29 wherein reagents are provided in dried form or freeze dried form.

31. The reaction vessel according to any one of claims 16 to 30 wherein an isothermal nucleic acid amplification reaction is used to detect or monitor the existence or quantity of a specific target in a sample.

32. A system according to any one of claims 1 to 15 further comprising at least one reaction vessel according to any one of claims 17 to 31.
33. A kit for performing concurrent isothermal nucleic acid amplifications, the kit comprising:
   a. a system according to any one of claims 1 to 15 or 32; and
   b. more than one reaction vessel configured to perform an isothermal nucleic acid amplification assay on a target sequence, each said reaction vessel being receivable in a reaction vessel receiving port of the system.

34. The kit according to claim 33 comprising a plurality of reaction vessels, wherein subsets of the reaction vessels are configured to perform isothermal nucleic acid amplification assays on different target sequences.

35. A method for identifying a pathogen in a sample using isothermal nucleic acid amplification comprising the steps of:
   a. collecting, and optionally preparing, a sample to be tested;
   b. selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an isothermal nucleic acid amplification assay on a target sequence;
   c. loading each reaction chamber with the sample;
   d. providing conditions suitable for performing isothermal nucleic acid amplification in reaction chambers concurrently;
   e. gathering information relating to amplified nucleic acid in reaction chambers;
   f. identifying pathogens present in the sample by processing the information gathered relating to amplified nucleic acid in a preprogramed central processing unit.

36. The method according to claim 35 wherein each reaction vessel identifies a different pathogen or different group of pathogens.

37. The method according to claim 35 or 36 wherein the plurality of reaction vessels are selected from a larger group of reaction vessels, wherein each reaction vessel is configured to perform at least one isothermal nucleic acid amplification on a target sequence.
38. The method of claim 37 wherein the group of reaction vessels is capable of identifying a plurality of different pathogens, preferably a group of pathogens greater in size than the number of reaction vessels in the selection.

39. The method of any one of claims 35 to 38 wherein isothermal nucleic acid amplifications are performed independently.

40. The method of any one of claims 35 to 39 wherein isothermal nucleic acid amplifications are performed concurrently.

41. A method for identifying the prevalence of a target pathogen, or pathogens, in a selection of different samples using isothermal nucleic acid amplification, comprising the steps of:
   a. collecting, and optionally preparing, a plurality of different samples to be tested;
   b. selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an isothermal nucleic acid amplification assay on a target sequence;
   c. loading each reaction vessel with a different sample;
   d. providing conditions suitable for performing isothermal nucleic acid amplifications to reaction vessels concurrently;
   e. gathering information relating to amplified nucleic acid in the reaction vessels; and
   f. identifying which of the samples contain the pathogen by processing the information gathered using a preprogramed central processing unit.

42. The method according to claim 41 wherein each of the reaction vessels is configured to perform an isothermal nucleic acid amplification on the same target sequence(s).

43. The method according to claim 40 or 42 wherein the reaction vessels are selected from a larger group of reaction vessels wherein a portion of reaction
vessels making up said group are configured to perform isothermal nucleic acid amplifications on different target sequences.

44. The method of any one of claims 41 to 43 wherein isothermal nucleic acid amplifications are performed independently.

45. The method of any one of claims 41 to 44 wherein isothermal nucleic acid amplifications are performed concurrently.

46. A method for performing concurrent isothermal nucleic acid amplification assays on target sequences comprising
   a. providing a first reaction vessel containing a first sample and a second reaction vessel containing a second sample, each reaction vessel being configured to perform an isothermal nucleic acid amplification assay on a target sequence;
   b. associating said reaction vessels with ports configured to gather information relating to amplified nucleic acid;
   c. identifying the reaction vessels and samples located within said reaction vessels;
   d. performing concurrent isothermal nucleic acid amplifications on target sequences present in the samples;
   e. gathering information relating to amplified nucleic acid from each reaction vessel;
   f. processing the information gathered relating to amplified nucleic acid with a central processing unit; and
   g. presenting information related to the amplified nucleic acid from each sample on a display interface.

47. A modular system for performing concurrent isothermal nucleic acid amplification assays on target sequences comprising:
   a. a first process module comprising a port configured to receive a reaction vessel, which is configured to perform an isothermal nucleic acid amplification assay on a target sequence contained in a sample when
received in said port, and a first detector for gathering information relating to amplified nucleic acid in said first reaction vessel;
b. at least one second process module comprising a second port configured to receive a second reaction vessel, which is configured to perform an isothermal nucleic acid amplification assay on target sequence contained in a sample when received in said second port, and a second detector for gathering information relating to amplified nucleic acid in said second reaction vessel;
c. means for identifying each reaction vessel and sample;
d. a control module comprising central processing unit configured to identify each reaction vessel and sample and to process information related to amplified nucleic acid in said identified reaction vessels; and
e. a user interface for presenting said processed information related to the amplified nucleic acid from said samples;

wherein the first process module, at least one second process module and control module detachably connect to one another.

48. The modular system according to claim 47 wherein the first process module, at least one second process module, the means for identifying each reaction vessel and sample, and user interface all communicate with the control module.

49. The modular system according to claim 47 or 48 wherein each process module comprises a reaction chamber, a fluid handling device, and a detector.

50. The modular system according to any one of claim 47 to 49 wherein the process module is configured to perform recombinase polymerase amplification of target sequences.

51. The modular system according to any one of claim 47 to 49 wherein a process module is configured to perform nicking and extension amplification reaction (NEAR) to target sequences.
52. The modular system according to any one of claims 47 to 51 wherein a process module is configured to amplify DNA within a sample without first thermally melting the DNA.

53. The modular system according to any one of claims 47 to 52 wherein the control module is programmed to send and receive data to the first and at least one second process modules, said data comprising initiating mixing sample(s) with reagents when sample(s) are present in reaction chambers; modulating the temperature of the reaction chambers to the desired isothermal set-point; monitor status of the reactions; actuate the detectors to acquire a measurement response to determine the presence or absence of target nucleic acid sequences in the one or more sample(s); format measurement response data and output result on user interface; and/or optionally communicate output results via communication port to a remote display terminal or a printer.

54. A system interface for connecting and controlling a plurality of process modules for performing isothermal nucleic acid amplification assays, comprising:
   a. a backbone connector comprising n+1 terminals to engage n+1 isothermal nucleic acid amplification process modules;
   b. a user interface;
   c. a control module for sending and receiving data to the n+1 isothermal nucleic acid amplification modules via the user interface and backbone connector;
   d. a common power source for providing power to the network and the n+1 connected isothermal nucleic acid amplification modules.

55. The system interface of claim 54 wherein the n+1 terminals of the backbone connector are specific to individual isothermal nucleic acid amplification modules.

56. The system interface of claim 54 wherein the n+1 terminals of the backbone connector are non-specific and may be connected to more than one isothermal nucleic acid amplification modules.
57. The system interface according to any one of claims 54 to 56 wherein the
control module manages flow of data between a user interface and the n+1
isothermal nucleic acid amplification modules.

58. The system interface according to any one of claims 54 to 57 further comprising
an identification reader, preferably a bar code scanner, a RFID scanner, a smart
card reader, or a camera.

59. The system interface of claim 58 wherein the identification reader receives data
relating to an individual from which a sample for analysis has been obtained.

60. The system interface of claim 58 or 59 wherein the identification reader receives
data relating to a reaction vessel for use in one of the process modules, and
wherein such data is used to configure the process module to perform a defined
isothermal nucleic acid amplification analysis on a sample inserted in the
process module.

61. The system interface according to any one claims 54 to 60 wherein n is an
integer greater than 1, preferably between 1 and 49, more preferably between
1 and 9, more preferably between 1 and 4.

62. A modular system for performing concurrent assays on at least one sample to
investigate the content thereof, the system comprising:

a. a first process module configured to receive at least one first reaction
   vessel configured to perform at least a first assay on a sample when
   received in said at least one first process module;

b. at least one second process module configured to receive at least one
   second reaction vessel configured to perform at least a second assay on
   a sample when received in said at least one second process module;

c. optional further process modules configured to receive at least one
   further reaction vessel configured to perform at least a further assay on
   a sample when received in said optional further process module;
d. at least one identification device for gathering information relating to the identity of each reaction vessel and/or each sample;

e. at least one detector per process module for gathering information relating to the output of each assay in each reaction vessel;

f. a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to provide information relating to the biological content of each sample; and

g. a display interface in communication with the central processing unit which presents information relating to the biological content of each sample.

63. The modular system according to claim 62 comprising a control module comprising i) a housing configured to receive at least the at least one first process module and the at least one second process module, and optionally to receive the optional further process module, within said housing, ii) the central processing unit, iii) the display interface, iv) optionally the identification device, and optionally iv) a power supply.

64. The modular system according to claim 63 wherein the process modules detachably connect to the control module.

65. The modular system according to claim 63 or claim 64 wherein the process modules are slidably receivable within the housing.

66. The modular system according to any one of claims 63 to 65 wherein housing is configured to receive process modules in a vertically stacked arrangement.

67. The modular system according to any one of claims 63 to 66 wherein the housing is configured to receive from about two to about ten process modules, preferably from about two to about six process modules.
68. The modular system according to any one of claims 63 to 67 wherein the housing is configured to receive process modules with different heights.

69. The modular system according to any one of claims 63 to 68 wherein first process module has a different height to the second process module.

70. The modular system according to claim 62 comprising a control module comprising the central processing unit, the display interface and optionally the at least one identification device, wherein the first and second process modules and the control module detachably connect to one another.

71. The modular system according to claim 70 further comprising a power module for powering the process modules and the control module, preferably wherein the power module comprises a mains power supply and/or a battery.

72. The modular system according to any one of claims 70 or 71 wherein the optional power module detachably connects to any one of the process modules.

73. The modular system according to any one of claims 70 to 72 wherein process modules, control module and, optional, power modules are detachably connectable in a vertical stack arrangement, preferably wherein the control module is locatable at a top of the stack, and the optional power module is locatable at a base of the stack.

74. The modular system according to any one of claims 63 to 73 wherein process modules are in wired or wireless communication with the control module.

75. The modular system according to any of claims 62 to 74 wherein the first and second ports are different, and/or wherein the first and second reaction vessels are different, and/or wherein the first and second assays are different.

76. The modular system according to claim 71 wherein the first and second assays are different and selected from isothermal nucleic acid amplification assays, such as NEAR and RPA; immunoassays; oral fluid drug screening assays; BGEM assay; INR assay.
77. The modular system according to any one of claims 62 or 72 or 73 wherein the reaction vessels are configured to perform a plurality of different assays on the same and/or different samples.

78. The modular system according to any one of claims 63 to 77 wherein the system is configured to receive and process reaction vessels independently.

79. The modular system according to any one of claims 63 to 78 wherein the central processing unit controls the processing of samples within the reaction vessels, preferably the heating of the samples; preferably test vessels are independently heated or heatable.

80. The modular system according to any one of claims 63 to 79 wherein the central processing unit controls the processing of one or more reaction vessels independently and/or concurrently.

81. The modular system according to any one of claims 63 to 80 wherein an individual reaction vessel is processed to yield a result in less than 15 minutes, less than 12 minutes, less than 10 minutes, less than 9 minutes, less than 8 minutes, less than seven minutes, less than six minutes, less than five minutes.

82. The modular system according to any one of claims 63 to 81 wherein an individual reaction vessel is processed to yield a result in at least about 5 minutes, at least about 7 minutes, at least about 8 minutes, at least about 9 minutes, at least about 10 minutes, at least about 12 minutes, at least about 15 minutes, at least about 20 minutes, at least about 30 minutes.

83. The modular system according to any one of claims 63 to 82 wherein a reaction vessel is identified by information presented on the reaction vessel, preferably wherein the information is presented in a bar code, a smart chip, an RFID tag or a key.

84. The modular system of claim 83 wherein the bar code is a linear bar code, a 2D bar code, a QR code, an alpha-numeric code.
85. The modular system of claim 83 wherein the information is acquired by direct contact between the identification device and the information presented on the reaction vessel.

86. The modular system of claim 83 wherein the coded information is acquired by indirect contact between the identification device and the information presented on the reaction vessel.

87. The modular system according to any one of claims 63 to 86 wherein the identification device comprises a coded information reader in communication with the central processing unit.

88. The modular system according to any one of claims 63 to 87 wherein the sample is a biological sample, a patient sample, a veterinary sample, or an environmental sample, the sample may be raw or optionally processed.

89. The modular system according to any one of claims 63 to 88 comprising a wired and/or wireless connection for transmitting the information relating amplified nucleic acid from the detector to the central processing unit.

90. The modular system according to any one of claims 63 to 89 wherein the reaction vessels comprise one or more sample chambers for containing the sample or samples to be processed.

91. A reaction vessel received in a modular system according to any one of claims 63 to 90.

92. The modular system according to any one of claims 63 to 90 comprising the at least one first reaction vessel and the at least one second reaction vessel and optionally at least one third reaction vessel.

93. A kit for performing concurrent assays on a sample, the kit comprising:

   a. a modular system according to any one of claims 63 to 90 or 92;
b. a plurality of first reaction vessels, each configured to perform a predetermined specific assay or assays on a sample and suitable for use in the at least one first process module;

c. a plurality of second reaction vessels, each configured to perform a predetermined specific assay or assays on a sample and suitable for use in the at least one second process module; and

d. optionally a plurality of further reaction vessels, each configured to perform a predetermined assay or assays on a sample and suitable for use in at least one of the optional further process modules.

94. The kit according to claim 94 wherein different each reaction vessel is configured to identify a different pathogen or group of pathogens or target analytes.

95. A method for identifying a pathogen or other target analyte in a sample using a plurality of different assays comprising the steps of:

a. collecting, and optionally preparing, a sample to be tested;

b. selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an assay;

c. selecting process modules for receiving and processing the selected reaction vessels, each process module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel;

d. gathering information relating to the identity of each reaction vessel and/or each sample;

e. functionally adjoining the process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or target analytes in the sample;

f. loading each reaction chamber with the sample;

g. providing conditions suitable for performing the assays;
h. gathering information relating to the assay output in the reaction chambers;
  i. identifying pathogens or analytes present in the sample by processing the information gathered using the central processing unit.

96. The method according to claim 95 wherein each reaction vessel identifies a different pathogen or different group of pathogens or other target analyte(s).

97. The method according to claim 95 or 96 wherein the plurality of reaction vessels is selected from a larger group of reaction vessels, wherein each reaction vessel is configured to perform at least one assay to identify at least one target pathogen or group of pathogens or other target analyte(s).

98. The method according to claim 97 wherein the group of reaction vessels is capable of identifying a plurality of different pathogens or groups of pathogens or other target analytes, preferably a number of pathogens or groups of pathogens or other target analytes greater in magnitude than the number of reaction vessels in the selection.

99. The method of any one of claims 95 to 98 wherein the assays are performed independently.

100. The method of any one of claims 95 to 99 wherein the assays are performed concurrently.

101. A method for identifying the prevalence of a target pathogen, or pathogens, or other target analyte(s), in a selection of different samples using a plurality of assays, comprising the steps of:
   a. collecting, and optionally preparing, a plurality of different samples to be tested;
   b. selecting a plurality of separate reaction vessels, each separate reaction vessel comprising at least one reaction chamber configured to perform an assay;
c. selecting process modules for receiving and processing the selected reaction vessels, each process module comprising at least one detector for gathering information relating to the output of each assay in each reaction vessel;

d. gathering information relating to the identity of each reaction vessel and/or each sample;

e. functionally adjoining the process modules to a control module comprising a central processing unit programmed to process the information gathered relating the output of each assay in each reaction vessel and the information gathered relating to the identity of each sample and/or reaction vessel to identify pathogens or other target analytes in the sample;

f. loading each reaction chamber with a different sample;

g. providing conditions suitable for performing the assays;

h. gathering information relating to the assay output in the reaction chambers;

i. identifying which of the samples contain the pathogen or other target analyte by processing the information gathered using the central processing unit.

102. The method according to claim 101 wherein each of the reaction vessels is configured to identify the same pathogen or group of pathogens or other target analyte(s).

103. The method according to claim 100 or 102 wherein the reaction vessels are selected from a larger group of reaction vessels wherein a portion of reaction vessels making up said group are configured to perform assays for identifying different target pathogens or other target analytes.

104. The method of any one of claims 101 to 103 wherein the assays are performed independently.

105. The method of any one of claims 101 to 104 wherein the assays are performed concurrently.
The method according to claim 35, 41, 46, 95, 101, or any claims dependent thereon, wherein the method is performed on a crude matrix sample.

5 106. A system, kit, device or method according to the examples, description and figures.
Collect a single sample to be tested.

Select plurality of different separate reaction vessels from a larger group of separate reaction vessels.

Load selected reaction vessels with the single sample.

Provide appropriate conditions for performing isothermal nucleic acid amplifications to reaction vessels concurrently.

Gather data relating to amplified nucleic acid.

Process data relating to amplified nucleic acid.

Identify pathogen(s) present in sample.

**FIG. 13**
Collect a plurality of different samples to be tested.

Select plurality of identical separate reaction vessels from a larger group containing multiple different separate reaction vessels.

Load each reaction vessel with a different sample.

Provide appropriate conditions for performing isothermal nucleic acid amplifications to reaction vessels concurrently.

Gather data relating to amplified nucleic acid.

Process data relating to amplified nucleic acid.

Identify which samples contain pathogen(s).

FIG. 14
Collect a single sample to be tested.

Select plurality of separate assay reaction vessels from a larger group of separate reaction vessels.

Select process modules for receiving and processing the reaction vessels.

Functionally adjoin the process modules to a control module.

Load selected reaction vessels with the single sample.

Provide appropriate conditions for performing the assays to the reaction vessels concurrently.

Gather data relating to the output of each assay.

Process data relating to the output of each assay.

Identify pathogen(s) or other target analyte(s) present in sample.
Collect a plurality of different samples to be tested.

Select plurality of identical separate assay reaction vessels from a larger group containing multiple different separate reaction vessel types.

Select a plurality of process modules for receiving and processing the reaction vessels.

Functionally adjoin the process modules to a control module.

Load each reaction vessel with a different sample.

Provide appropriate conditions for performing the selected assay to the reaction vessels concurrently.

Gather data relating to the assay output.

Process data relating to the assay output.

Identify which samples contain identified pathogen(s) or target analyte(s).

FIG. 22
## INTERNATIONAL SEARCH REPORT

**International application No**

PCT/EP2016/064085

### A. CLASSIFICATION OF SUBJECT MATTER

**INV.** C12Q1/68  
**ADD.** G01N35/00

According to International Patent Classification (IPC) or to both national classification and IPC

### B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

C12Q  G01N  A61B

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where predictable, search terms used)

EPO-Internal, WPI Data

### C. DOCUMENTS CONSIDERED TO BE RELEVANT

<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>paragraphs [0032], [0036]; figure 4</td>
<td>47-53, 62-90, 92-105</td>
</tr>
<tr>
<td></td>
<td>paragraphs [0017], [0021] - [0026], [0055] - [0064]; figures 1-3</td>
<td>47-53, 62-90, 92-105</td>
</tr>
<tr>
<td></td>
<td>paragraphs [0395], [0397], [0447], [0484], [0511] - [0514]; figures 5,6</td>
<td></td>
</tr>
</tbody>
</table>

**X** Further documents are listed in the continuation of Box C.  

**X** See patent family annex.

* Special categories of cited documents:

**A** document defining the general state of the art which is not considered to be of particular relevance

**E** earlier application or patent but published on or after the international filing date

**L** document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

**O** document referring to an oral disclosure, use, exhibition or other means

**P** document published prior to the international filing date but later than the priority date claimed

**X** later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

**X** document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

**Y** document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

**A** document member of the same patent family

**Date of the actual completion of the international search**

19 August 2016

**Date of mailing of the international search report**

31/10/2016

**Name and mailing address of the ISA/IEA**

European Patent Office, P.B. 5618 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040,  
Fax. (+31-70) 340-3016

**Authorized officer**

van Lith, Joris
<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>WO 02/43864 A2 (CLINICAL MICRO SENSORS INC [US]; TERBRUEGGEN ROBERT [US]) 6 June 2002 (2002-06-06) page 2, line 27 - page 5, line 2; figures 1-7</td>
<td>1,46,47, 62,101</td>
</tr>
</tbody>
</table>
**INTERNATIONAL SEARCH REPORT**

### Box No. II  Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. □ Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely:

2. □ Claims Nos.: because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. □ Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

### Box No. III  Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

> see additional sheet

1. □ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. □ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of additional fees.

3. □ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. □ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

   1-15, 32-34, 46-53, 62-90, 92-106

**Remark on Protest**

- □ The additional search fees were accompanied by the applicant’s protest and, where applicable, the payment of a protest fee.
- □ The additional search fees were accompanied by the applicant’s protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- □ No protest accompanied the payment of additional search fees.
This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

   Invention 1 is directed to a system comprising multiple ports for receiving reaction vessels
   ---

2. claims: 16-31, 91
   Invention 2 is directed to a reaction vessel.
   ---

3. claims: 35-45
   Invention 3 is directed to a method for identifying a pathogen.
   ---

4. claims: 54-61
   Invention 4 is directed to a system interface for controlling a plurality of process modules for performing isothermal nucleic acid amplification assays.
   ---
<table>
<thead>
<tr>
<th>Patent document cited in search report</th>
<th>Publication date</th>
<th>Patent family member(s)</th>
<th>Publication date</th>
</tr>
</thead>
<tbody>
<tr>
<td>US 2013078736 A1</td>
<td>28-03-2013</td>
<td>AU 2012311434 A1</td>
<td>27-03-2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td>AU 2016209920 A1</td>
<td>03-03-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2849193 A1</td>
<td>28-03-2013</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN 103945941 A</td>
<td>23-07-2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN 105170202 A</td>
<td>23-12-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CN 105181390 A</td>
<td>23-12-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 2758172 A2</td>
<td>30-07-2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HK 1200399 A1</td>
<td>27-11-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP 2014528075 A</td>
<td>23-10-2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2013078736 A1</td>
<td>28-03-2013</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016288116 A1</td>
<td>06-10-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WD 2013041713 A2</td>
<td>28-03-2013</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP W02014020977 A1 A</td>
<td>21-07-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SG 11201500649U A</td>
<td>30-03-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2015251184 A1</td>
<td>10-09-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WD 2014020977 A1</td>
<td>06-02-2014</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2015338428 A1</td>
<td>26-11-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2015368717 A1</td>
<td>24-12-2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016003823 A1</td>
<td>07-01-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016011215 A1</td>
<td>14-01-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016011225 A1</td>
<td>14-01-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016025760 A1</td>
<td>28-01-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016025763 A1</td>
<td>28-01-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016033544 A1</td>
<td>04-02-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016069921 A1</td>
<td>10-03-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016169880 A1</td>
<td>15-06-2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2016169923 A1</td>
<td>16-06-2016</td>
</tr>
<tr>
<td>US 2004248087 A1</td>
<td>09-12-2004</td>
<td>AU 6376698 A</td>
<td>05-11-1998</td>
</tr>
<tr>
<td></td>
<td></td>
<td>BR 9801572 A</td>
<td>29-06-1999</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2230967 A1</td>
<td>02-11-1998</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 0875584 A2</td>
<td>04-11-1998</td>
</tr>
<tr>
<td></td>
<td></td>
<td>ES 23500038 T3</td>
<td>17-01-2011</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP 4526609 B2</td>
<td>18-08-2010</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP H10304890 A</td>
<td>17-11-1998</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 6300668 B1</td>
<td>09-10-2001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 6528632 B1</td>
<td>04-03-2003</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 6558901 B1</td>
<td>06-05-2003</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 6586234 B1</td>
<td>01-07-2003</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2004248087 A1</td>
<td>09-12-2004</td>
</tr>
<tr>
<td></td>
<td></td>
<td>US 2008240985 A1</td>
<td>02-10-2008</td>
</tr>
<tr>
<td>EP 1541237 A2</td>
<td>15-06-2005</td>
<td>NONE</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>CA 2427669 A1</td>
<td>06-06-2002</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EP 1331999 A2</td>
<td>06-08-2003</td>
</tr>
<tr>
<td></td>
<td></td>
<td>JP 2004515231 A</td>
<td>27-05-2004</td>
</tr>
<tr>
<td></td>
<td></td>
<td>WO 0243864 A2</td>
<td>06-06-2002</td>
</tr>
</tbody>
</table>