(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
24 March 2011 (24.03.2011)

PCT

(10) International Publication Number
WO 2011/032941 A1

(51) International Patent Classification:
G01N 33/543 (2006.01) *G01N 35/00* (2006.01)
B01L 3/00 (2006.01) *B04B 5/04* (2006.01)

(21) International Application Number:
PCT/EP2010/063477

(22) International Filing Date:
14 September 2010 (14.09.2010)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
09170833.9 21 September 2009 (21.09.2009) EP

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(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— *as to the identity of the inventor (Rule 4.17(i))*

[Continued on next page]

(54) Title: METHOD FOR CARRYING OUT REACTIONS IN AN ANALYTICAL DEVICE

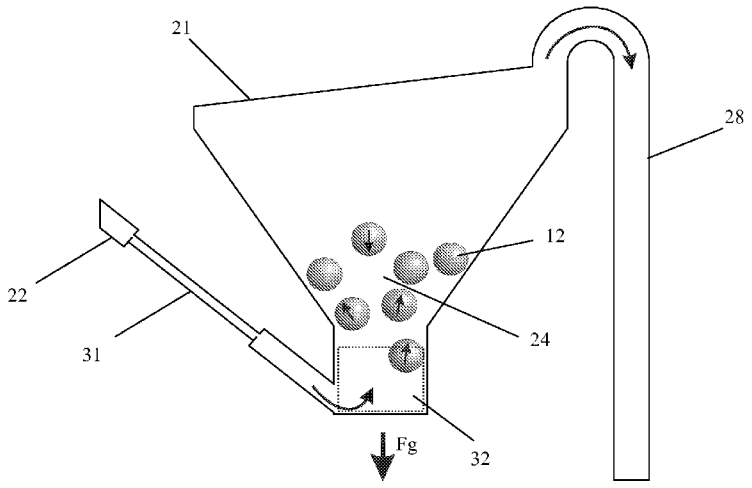

Fig. 2

(57) Abstract: Method and system for carrying out heterogeneous chemical or biological reactions, the method comprising the steps of providing an analytical device comprising at least one liquid processing unit, the liquid processing unit comprising at least one reaction chamber, and at least a first inlet channel in fluid communication with the reaction chamber, supplying to the reaction chamber via the first inlet channel or a second inlet channel analyte capturing particles, supplying to the reaction chamber via the first inlet channel or the second inlet channel a liquid sample containing at least an analyte of interest, confining by an equilibrium of forces the analyte capturing particles in a particle rearrangement zone comprised in the reaction chamber, said forces comprising a drag force Fd generated by liquids flowing and a counter-oriented force Fg, capturing analytes present in the liquid sample with the particles in the particle rearrangement zone.

WO 2011/032941 A1

Published:

— *with international search report (Art. 21(3))*

METHOD FOR CARRYING OUT REACTIONS IN AN ANALYTICAL DEVICE

TECHNICAL FIELD

The present invention is in the field of in-vitro diagnostics and concerns a method for carrying out heterogeneous chemical or biological reactions involving the use of analyte capturing particles.

BACKGROUND OF THE INVENTION

In the field of in-vitro diagnostics there is a strong need for the automated heterogeneous assay of chemical and biological samples. Due to several advantages, among which low sample and reagent requirement, the possibility to integrate several assay steps into a single device, increased reaction kinetics, fast analysis times and hence high throughput, in recent years, many efforts have been made to develop microfluidic systems.

Packed bed columns, built up as a pile of beads, are common knowledge and used for e.g. chromatography in chemical analysis since decades. Also in diagnostic applications the use of porous frit structures is known in the art. For example immunoassays on a rotating disc where liquids flow through a packed bed column, driven by centrifugal force, have been disclosed. Different ways of building columns with beads or particles in miniaturized systems are known. A conventional way is that to use slurries from which a fixed structure is obtained. Typically a bead-liquid emulsion or suspension is flown through, along or over a chamber in which the beads are trapped. This trap can be either a small passage, such as a shallow slit, a porous material like a fleece or filter, e.g. a sintered porous material, or larger beads. Another method is to trap the particles in a magnetic field. Typically all these methods have in common that the particles are trapped in a forward flow direction. This means that the flow-resistance is fixed or increased when liquid is flown through. Due to an increased packing grade while trapping the particles, the column flow resistance will increase, possibly leading to clogging. When the column is used to bind molecules flowing through, this effect can be even more severe. In case the beads are trapped in a field, like for instance magnetic, the drag forces may exceed the trapping forces on the beads, resulting in bead loss. When single particles are packed in a geometrical way via a narrowed passage, the obtained pore-size depends on the bead-size. Because larger particles may not provide suf-

ficient binding capacity, smaller particles are preferred but they will yield smaller pores and thus a higher flow resistance. As a consequence, there is a practical limitation of what bead-size can be used for a given flow pressure range. This can be a problem when for instance using centrifugation for driving the flow as the obtainable pressures are small. Once a column is clogged, it may be very difficult to recover it.

Unlike flow-through configurations, US 2007/0183935 discloses a device comprising a pair of bellows pumps configured for moving a fluid sample and particles in suspension through an aperture between paired bellows pump mixing chambers.

It is an object of the present invention to provide methods of carrying out heterogeneous chemical or biological reactions involving the use of analyte capturing particles, which enables efficient capturing of analytes while avoiding high flow resistance and the risk of clogging.
This is achieved by confining the analyte capturing particles in a particle rearrangement zone, in which, due to equilibrium of forces, the particles are able to rearrange themselves, but from which the particles are unable to escape.
Another advantage of the present invention is that mixing with reagents is more efficient thus increasing the kinetics of reaction, reducing further the time of the assay and the consumption of samples and reagents.
Another advantage is that the analytical device can be generic, simple and cost efficient, wherein the particles can be introduced in the form of a reagent at the moment of the assay without the need to pre-pack a particle column in advance or during manufacturing. In an alternative embodiment, the particles can be applied, preferably as a suspension, directly into the reaction chamber of the analytical device and dried therein during the manufacturing of the analytical device. The advantage of this embodiment is that the particles are already present in the analytical device and will be resuspended by a liquid, preferably by the sample itself, automatically during test performance without additional steps of reagent admixture. Moreover various types and sizes of particles can be used which enormously increases the flexibility of the assay and the device.

GENERAL DESCRIPTION OF THE INVENTION

The present invention relates to a method for carrying out heterogeneous chemical or biological reactions comprising the steps of:
a) providing an analytical device comprising at least one liquid processing unit, the liquid processing unit comprising

- at least one reaction chamber and at least a first inlet channel in fluid communication with the reaction chamber,

b) supplying to the reaction chamber via the first inlet channel or a second inlet channel analyte capturing particles, or providing analyte capturing particles in the reaction chamber, preferably by a analyte capturing particles pre-loading step during manufacturing of the analytical device,

c) supplying to the reaction chamber via the first inlet channel or the second inlet channel a liquid sample containing at least an analyte of interest,

d) confining by an equilibrium of forces the analyte capturing particles in a particle rearrangement zone comprised in the reaction chamber, said forces comprising a drag force Fd generated by liquids flowing through the first inlet channel and a counter-oriented force Fg,

e) capturing analytes present in the liquid sample with the particles in the particle rearrangement zone.

The analytical device according to the present invention may be used in chemistry like in in-vitro diagnostics, and is adapted to carry out various assay operations comprising mixing liquids as well as detecting analytes contained in liquid samples. It may be for example used for diagnostic assays like e.g. clinical chemistry assays and immunoassays. Typical diagnostic assays include for example the qualitative and/or quantitative analysis of analytes such as albumin, ALP, Alanine Aminotransferase, Ammonia, Amylase, Aspartat Aminotransferase, Bicarbonate, Bilirubin, Calcium, Cardiac Markers, Cholesterol, Creatinine Kinase, D-Dimer, Ethanol, g-Glutamyltransferase, Glucose, HBA1c, HDL-Cholesterol, Iron, Lactate, Lactate Dehydrogenase, LDL-Cholesterol, Lipase, Magnesium, Phosphorus inorganic, Potassium, Sodium, Total Protein, Triglycerides, UREA, Uric Acid. The list is of course not exhaustive.

According to the present invention, the recited liquids may be samples and reagents. Samples are liquid solutions in which one or more analytes of interest can be potentially found. Samples may comprise chemical analytes, e.g. organic chemicals, and the analytical device can be adapted to carry out one or more chemical assays, e.g. a drug interaction screening, an environmental analysis, the identification of organic substances, synthesis, etc... Samples can be also biological as e.g. body fluids like blood, serum, urine, milk, saliva, cerebrospinal fluid, etc...and may comprise chemical as well as biological analytes, such as nucleic acids, proteins, peptides, lipids, metabolites, etc...

According to the present invention, the term reagent is used to indicate any liquid, e.g. a solvent or chemical solution, which needs to be mixed with a sample and/or other reagent in order e.g. for a reaction to occur, or to enable detection. A reagent can be for example another sample interacting with a first sample. A reagent can be also a diluting liquid, it may comprise an organic solvent, a detergent, it may be a buffer or a solution for performing washing steps. A reagent in the more strict sense of the term may be a liquid solution containing a reactant, typically a compound or agent capable e.g. of binding to or transforming one or more analytes present in a sample, eventually producing a detectable result. Examples of reactants are enzymes, enzyme substrates, conjugated dyes, protein-binding molecules, nucleic acid binding molecules, antibodies, chelating agents, promoters, inhibitors, epitopes, antigens, catalysts, etc... Optionally dry reagents may be present in the analytical device and be dissolved by a sample, another reagent or a diluting liquid.

Typically, reagents are mixed with samples and the assay is a heterogeneous reaction making use of a solid support. According to the present invention, the solid support consists of particles susceptible to a force such as a drag force and a field force, and which remain corpuscular, i.e. are suspendable but not dissolvable in the liquid they come in contact with. Said particles are typically analyte capturing particles, wherein the term capturing indicates the ability of the particles, directly or indirectly, e.g. via a surface coating, to bind or retain at least temporarily at least one analyte contained in the sample and/or to interact in a specific or selective manner with said analytes.

The following are examples of heterogeneous reactions:

- Heterogeneous immunoassay.
- Catalysis, where the particles may have a catalytic surface.
- Capturing and purification of nucleic acids.
- Capturing of other biological material such as cells or viruses.
- Affinity chromatography.
- Chemical solid phase extraction.

Said particles are preferably embodied as suspendable beads or micro-spheres with a diameter e.g. in the micrometer range, e.g. comprised between 0.1 and 20 micrometer, preferably between 1 and 5 micrometer. In applications requiring a high sensitivity, the use of smaller particles is preferred, because such smaller particles comprise a higher surface-to volume ratio which is beneficial for a higher sensitivity of an assay. In alternative embodiments, particles of different density and different analyte capturing properties can be applied into the reaction chamber. The use of analyte capturing particles with different

densities and different analyte capturing properties allows a simultaneous detection of multiple analytes by their respective analyte capturing particles, because the different analyte capturing particles can be sorted according to their densities in different zones of the detection compartment (e.g. by centrifugal forces). Also, by size, shape and density, the drag and field forces on the beads can be "tuned" or tailor-made to the specific needs. By surface roughness and porosity, the surface-to-volume-ratio can be adapted. Commercially available beads such as latex beads or magnetic beads provided with immobilized reactants, or with binding elements to which reactants can be immobilized, are examples of suitable particles. In some embodiments, the use of transparent particles is beneficial, because the use of at least partially transparent particles allows an optical detection also in depth (i.e. in all 3 spatial dimensions) because the upper particle layers of a packed particle bed will not shadow the layers below. This results in an increased signal intensity and increased sensitivity.

The analyte capturing particles can be supplied to the analytical device in the form of a liquid reagent, e.g. a liquid suspension, or are already provided in the reaction chamber, preferably by an analyte capturing particles pre-loading step during manufacturing of the analytical device, and are resuspended by a liquid, preferably by the sample itself, automatically during test performance without additional steps of reagent admixture.

According to the present invention, the analytical device has a device body comprising at least one liquid processing unit. According to one embodiment the device body is a carrier which comprises one or more liquid processing units. The device body and the liquid processing units may be separate entities joined with each other at the moment of use. In this case the device body could be made of a rigid material, e.g. metal, glass or ceramics, or a plastic material, e.g. injection molded, and has functional features such as e.g. compartments to receive liquid, processing units, alignment pins and/or holes, clamps, levers, or screws to fix the liquid processing units. The device body may have holes enabling optical detection or may be transparent.

According to a preferred embodiment the device body and the at least one liquid processing unit form one integral piece, made e.g. of a plastic material, preferably injection molded. The device body is preferably at least partially transparent. According to a preferred embodiment the device body is disposable.

The device body has preferably a disc-like shape, preferably round, e.g. with the footprint of a compact disc (CD).

A liquid processing unit is either a separate element that can be coupled to the device body, or an integral part of the device body, comprising interconnected microfluidic structures by which it is possible to achieve miniaturization and integration of the various assay operations. The term integral is here used to indicate that the liquid processing unit is at least partially built in the device body at the moment of production and is not separable from the device body.

A liquid processing unit comprises at least two layers, one substrate layer and one cover layer. The microfluidic structures are created preferably on the upper surface of the substrate and sealed from the top with the cover layer. According to one embodiment, the substrate layer is the device body. According to another embodiment, the substrate layer is a separate element that can be coupled to the device body. The cover layer can be made of the same material as the substrate layer or of a different material such as e.g. a thin polymeric foil, preferably transparent. A preferred way of achieving sealing between the substrate layer and the cover layer is to use bonding technologies, such as thermal bonding, gluing, injection molding, e.g. two-component injection molding, etc.... The sealing preferably occurs at the moment of production. It may however occur before use. Terms like upper and top are here used as relative and not absolute. The position of substrate layer and cover layer can be for example reversed. The cover layer comprises preferably holes or access ports to enable the access of liquids such as samples, reagents and/or air to the microfluidic structures.

According to a preferred embodiment a plurality of liquid processing units are symmetrically arranged around a central axis of rotation of the device body.

According to the present invention, the liquid processing unit comprises at least one reaction chamber, for mixing at least one liquid with analyte capturing particles. The reaction chamber is a microfluidic structure defined as a cavity in or between the substrate layer and the cover layer, defining a lower wall and upper wall respectively, and delimited by side walls. The volume of the reaction chamber defines the maximum volume of reaction mixture. Said volume can be in the microliter and milliliter range, e.g. 1 microliter up to several milliliters, typically below 1 mL.

The liquid processing unit further comprises at least one inlet channel in fluid communication with the reaction chamber for delivering at least one liquid to the reaction chamber.

According to a preferred embodiment the liquid processing unit comprises at least one inlet chamber in fluid communication with the reaction chamber via at least a first inlet channel.

Different liquid processing units may be partially interconnected between them, e.g. one access port or inlet channel might be in common to more than one liquid processing unit.
At least a part of the reaction chamber is designed as a particle rearrangement zone, i.e. a zone in which the particles are able to rearrange themselves, but from which the particles are unable to escape, at least temporarily.
In the embodiment of a flow-through variant, the particle rearrangement zone is defined by equilibrium of forces, said forces comprising a drag force generated by liquids flowing through said first inlet channel and a counter-oriented force. Particles may escape the particle rearrangement zone or may stop rearranging themselves if the equilibrium between the two types of forces is lost, e.g. if one type of force is reduced or increased compared to the other or one is removed.

A drag force is a hydrodynamic force dependent on flow velocity generated by liquid flowing, e.g. by a liquid flowing into the reaction chamber via said at least one first inlet channel, and acting on the particles suspended in the liquid in the direction of the liquid flow relative to the particles. Drag forces can also be induced by Euler forces (which are induced by accelerations of the analytical device) in combination with appropriate chamber designs (see shake-mode variant below).

A counter-oriented force is a force acting on the analyte capturing particles induced by a field chosen from the group of gravitational field, magnetic field, electromagnetic field, electrostatic field, acceleration field, comprising centrifugal, translational, rotational acceleration fields.

In other words, the drag force and the counter-oriented force are opposite to each other.
As the liquid flow tends to drag particles in the flow direction, the field-induced force tends to move the particles in the opposite direction against the liquid flow.
By regulating the field-induced force and/or the liquid flow velocity, the boundaries of the particle rearrangement zone can be varied. For example, if the field-induced force is increased and the drag force is decreased, the particle rearrangement zone becomes smaller, i.e. the particles are less dispersed and rearrange themselves only in a limited volume or part of the reaction chamber. If the drag force is stopped, the particles may eventually sediment or agglomerate with very little or no rearrangement occurring any longer. This situation may occur temporarily, e.g. before a new liquid addition and it may be preferred at the end of the reaction when detection is eventually performed, because such an agglomeration of the particles results in an increased signal intensity and sensitivity.

If, on the contrary, the counter-oriented force is decreased and the drag force increased, the particle rearrangement zone becomes larger, i.e. the particles are more dispersed and rearrange themselves in a larger volume or part of the reaction chamber. If the counter-oriented force is further decreased, the particles may eventually be dragged out of the reaction chamber.

So, by regulating drag and counter-oriented forces the effective particle concentration, diffusion distances and flow resistance can be determined and adjusted.

In an alternative embodiment to the flow-through variant described above, the shake-mode variant, the arrangement of the particles is achieved by the interaction of a field force with a counter-orientated drag force which is induced by Euler forces. The use of Euler forces in microfluidic structures is well-known and used in different embodiments, most preferably for mixing purposes in a shake-mode as described in e.g. "Unidirectional shake-mode for mixing highly wetting fluids on centrifugal platforms"; S. Lutz et al.; 12th International Conference on Miniaturized Systems for Chemistry and Life Sciences, October 12 – 16, 2008, San Diego. Euler forces result from an acceleration of the analytical device and have effects both on the suspended particles and particularly on the liquid in the reaction chamber. Euler forces are occurring in a direction perpendicular to the field force (centrifugal force). Depending on the geometric design of the reaction chamber, the resulting fluid flows can be occur also in other directions, because the walls of the reaction chamber can deflect the originally perpendicular flow into different directions.

Caused by the alternating acceleration of the analytical device (e.g. by a see-saw movement of the analytical device) and the alternating Euler forces, a liquid flow is induced which flows alternating from one side of the reaction chamber to the other side of the reaction chamber und which flows through the particle arrangement at each change of acceleration of the analytical device. Compared to the alternative embodiment of a flow-through variant, a shake-mode can also cause a kind of pump functionality which moves a liquid multiple times through a particle arrangement. Therefore, all the advantages and options described in connection with the flow-through variant are also applicable for the shake-mode variant.

If the analytical device is designed as a centrifugal test device, the field force can be generated by a fast rotational movement, preferably with a constant velocity. In this embodiment, the field force is the centrifugal force. The counter-oriented drag forces induced by Euler forces can be generated by rapid changes of the rotation direction (shake-mode).

For the shake-mode, the particles have preferably a density which is slightly larger than the density of the surrounding liquid (e.g. sample). This allows both a sedimentation and ag-

glomeration of the particles for detection purposes (which increases the signal intensity and sensitivity) and also an improved resuspension of the particles in the shake-mode variant. The „pumping“ of the liquid through the particle arrangement occurs preferably in a shake-mode with low final rotational velocities and high rotational accelerations. This results in large Euler forces (resulting in large drag forces) in combination with a loosened particle arrangement and therefore in an optimized flow-through of the liquid relative to the particle arrangement which increases the binding efficiency.

By using the shake-mode variant, the field force is a centrifugal force which varies strongly from zero at the beginning of the acceleration (zero rotational speed) to maximum at the end of the acceleration phase (maximum rotational speed). This means that initially the beads gets dragged with the flow but once the centrifugal force becomes larger than the drag forces at higher rotational speeds, the beads but also the fluid flows backwards thereby confining the beads in a capturing zone and levelling the fluids at both sides of the capturing zone. (The capturing zone is located at the part of the reaction chamber which is most distant from the rotational axis. In the described embodiments, the capturing zone is located within the connecting section 71c.) Since the beads have a larger density than the fluid, they are stronger influenced by the centrifugal forces than the fluid. Hence the beads are more confined to the capturing zone than the fluid, resulting in a bed of beds that is flown through in alternating directions by the fluid. By defining the rotational accelerations and speed profiles, the effect of the pumping action due to Euler forces and bead-capturing due to centrifugal forces can be controlled.

For the agglomeration of the particles, e.g. for detection purposes, a high and constant rotational force is preferred. This results in no Euler, but high centrifugal forces forcing the beads (which have a higher density than the liquid) to agglomerate on the side of the reaction chamber which is most distant from the rotation axis.

With both variants, the flow-through variant and the shake mode variant, and due to the above mentioned methods for loosening and agglomerating the particles, an efficient mass transport (resulting in an improved binding and washing efficiency) can be combined with a agglomerated and compact arrangement of the particles for detection (resulting in increased signal intensity and sensitivity).

The method of the present invention may further comprise the steps of:

f) supplying to the reaction chamber via the first inlet channel a first liquid reagent comprising signal generating conjugates for binding to analytes captured by the analyte cap-

turing particles or providing a reagent comprising signal generating conjugates for binding to analytes captured by the analyte capturing particles in the reaction chamber (21, 71), preferably by a reagent pre-loading step during manufacturing of the analytical device,

g) supplying to the reaction chamber via the first inlet channel a second liquid reagent for washing unbound signal generating conjugates out of the particle rearrangement zone,
h) detecting the signal generated by the conjugate-bound analytes captured by the particles.

For efficient reactions and/or efficient washing, i.e. fast reaction kinetics and low volume consumption, it is advantageous that the analyte capturing particles rearrange themselves. In the particle rearrangement zone the particles are in continuous movement, i.e. they continuously change position within the liquid, thus resulting in improved mixing. This means higher chance and shorter time for the particles to find an analyte to capture, for the bound analytes or reagents to find a reactant in the liquid solution and for the bound-free reagents to be more easily washed away after the reaction, while using smaller volumes.

In defining the boundaries of the particle rearrangement zone, the geometry of the reaction chamber may play an important role.

According to preferred embodiment of a flow-through variant, the reaction chamber, at least in correspondence of the particle rearrangement zone, has a tapered or funnel structure with diverging walls in the flow direction. This geometry gradually reduces the downstream directional flow velocity of liquids flowing in the reaction chamber via said first inlet channel, wherein said first inlet channel is fluidically connected to the narrower edge or corner of the tapered structure or bottom side of the funnel. As the width of the reaction chamber increases the flow velocity and hence the drag force acting on the particles decreases until the drag force is outbalanced by the counter-oriented force, which prevents the particles to move further downstream. The method may therefore comprise the step of reducing the directional flow velocity of liquids entering the reaction chamber via said first inlet channel by designing the reaction chamber with a tapered shape.

The method of the present invention may comprise the step of splitting liquids flowing through the reaction chamber in multiple streams. This has the effect to improve the flow profile, e.g. to flatten the flow profile, which normally tends to be parabolic, or to have the particles more distributed in the centre of the reaction chamber and less on the outer walls. According to another embodiment the method may comprise the step of dividing the reaction chamber in a serially arranged set of diffuser-like compartments. Diffuser-like com-

partments in series may be used to trap a fraction, e.g. size dependent, or a part of the particles, and/or be used for different reaction steps.

Also, the flow velocity can be controlled by varying the size of the inlet channel and/or designing the inlet channel with a flow resistor, e.g. a restriction of the cross-section inducing hydraulic resistance to the flow and thus reducing the flow velocity. The method of the present invention may thus comprise the step of controlling the flow velocity of liquids through said first channel via a flow resistor.

Ideally, thus the entire reaction chamber or at least the part being filled with liquid is a particle rearrangement zone, so that the particles are distributed throughout the liquid. Typically, the particle rearrangement zone occupies less than 50% of the volume of the reaction chamber, in order to stay within safety margins in case of drag or counter-oriented force fluctuations.

According to preferred embodiment of a shake-mode variant, the reaction chamber is divided by a central element into 2 sub-chambers which are connected by a connecting section which connects the two sub-chambers at their ends which are distant from a rotational axis. The central element works as a barrier and causes a flow of liquid through the connecting section from one sub-chamber into the other sub-chamber in the shake-mode. During the shake-mode, the liquid is alternately “pumped” from one sub-chamber into the other sub-chamber. Also the particles are arranged in the connecting section (and optionally also in the adjacent parts of the respective sub-chambers), due to their higher density. The pumping movement forced by the Euler forces in the shake-mode causes a multiple flow-through of the liquid through the particle arrangement in this section. These multiple liquid movements relative to the particles result in an increased binding efficiency of the analyte molecules within the liquid to their respective binding partners which are immobilized onto the surfaces of the particles.

Like the preferred embodiments of the flow-trough variants, also preferred embodiments of the sub-chambers of a shake-mode variant have a geometric structure with diverging walls in a flow direction directed to the rotational axis. In a preferred embodiment, this tapering in direction to the connecting section occurs in all spatial directions, i.e. also the height of the reagent chamber decreases in direction to the rotational axis-distant connecting section. Because this connecting section is the most distant part of the reaction chamber in relation to the rotational axis, an agglomeration of the particles by centrifugal forces will occur in this section. This agglomeration of particles by centrifugal forces is supported by this preferred

geometric design of the reaction chamber, because the decrease of the geometric dimensions towards this part of the reaction chamber causes additional capillary forces which are beneficial to keep the particles in this part of the reaction chamber, even at relatively low rotational speeds. All together, such a geometric design is beneficial for an agglomeration of the particles in this part of the reaction chamber and therefore in a preferred embodiment this connecting section (and optionally the adjacent parts of the respective sub-chambers) will be also used as a detection chamber in which the optical signal of the agglomerated particles can be detected.

The reaction chamber may comprise one or more inlet channels (e.g. for sample fluids, liquid reagents or washing solutions). Preferably, these inlet channels are located on the opposite side of the reaction chamber in relation to the connecting section, i.e. close to the rotational axis.

The reaction chamber may comprise also one (or more) outlet channels. In a preferred embodiment of a shake-mode variant, the outlet channel is located slightly above the particle arrangement zone. This allows an almost entire exchange or removal of liquids within this zone without a loss of particles which results in a very efficient washing characteristic of such an embodiment.

In a preferred embodiment of the outlet channel, the outlet channel is designed in form of a microfluidic siphon which comprises valving functionalities and allows a control of the chamber emptying process by preventing the emptying of the reaction chamber during the shake-mode process. The geometric design of such microfluidic siphons is well-known (e.g. US 6235531).

According to a preferred embodiment the liquid processing unit has at least two inlet channels in fluid communication with the reaction chamber, wherein the second inlet channel is used e.g. to introduce at least the particles, and the first channel is used to introduce the other liquids, which upon entering the reaction chamber exercise a drag force on the particles introduced via the second channel. This may prevent e.g. clogging of the first inlet channel, especially if the cross-section is small or if it comprises a flow resistor.

In an alternative embodiment, the particles can be applied, preferably as a suspension, directly into the reaction chamber of the analytical device and dried therein during the manufacturing of the analytical device. For these purposes, the reagent chamber may comprise in a preferred embodiment recesses which allow a controlled and defined application and drying of particles or also other reagents directly into the reaction chamber.

According to a preferred embodiment the liquid processing unit comprises at least one outlet channel.

The liquid processing unit may further comprise at least one waste chamber in fluid communication with the reaction chamber via the at least one outlet channel for receiving liquids flowing out of the reaction chamber. The waste chamber is typically larger than the reaction chamber in order to accommodate volumes of liquids which sequentially and/or continuously flow through the reaction chamber.

The method of the present invention may thus comprise the step of inducing liquids to flow out of the reaction chamber into a waste chamber in fluid communication with the reaction chamber via an outlet channel, while the particles are retained in the reaction chamber.

One waste chamber may receive liquid from more reaction chambers from the same or different liquid processing units.

The liquid processing unit may further comprise a liquid dosing chamber for delivering a defined volume of liquid, e.g. sample, to the reaction chamber. A liquid dosing chamber is a microfluidic structure defined as a cavity in or between the substrate layer and the cover layer, the volume of which defines the volume of liquid to be used in the assay once it has been filled. Said volume is typically below 1 microliter, e.g. about 200 nanoliters. The liquid dosing chamber has preferably an elongated shape and has at least two microchannels connected to it: a third liquid inlet channel allowing liquid to fill the liquid dosing chamber; one liquid decanting channel, defining where the liquid dosing chamber starts and the liquid inlet channel ends, and allowing excess liquid to be guided to a waste chamber. At about the opposite side, the sample dosing chamber comprises a microfluidic valve, e.g. a geometric or hydrophobic valve based on changes of the geometrical surface characteristics and surface energy. One way of realizing a geometric valve is by a restricted conduit ending blunt at the inner edge of a larger channel or chamber. Maintaining the driving force, typically a counter-oriented force, below that required to break the energy barrier of the valve will cause the liquid to stop at this position and any excess to be deviated to the decanting channel characterized by having a barrier energy lower than that of the valve.

The method of the present invention may thus comprise the step of dosing defined volumes of liquids to be delivered to the reaction chamber via at least one liquid dosing chamber in fluid communication with the reaction chamber via at least said first inlet channel.

Bypass channels in fluid communication with the first inlet channel may me used to introduce other liquids, including e.g. diluting liquids.

According to the present invention the reaction chamber may also serve as detection chamber. This means that the presence and/or quantization of captured analytes is determined directly in the reaction chamber after or during the mixing between liquids and the analyte capturing particles. Detection is typically optical detection, e.g. based on photometric methods such as absorbance measurement, turbidimetry, luminescence, bioluminescence, chemiluminescence, fluorescence, phosphorescence. Therefore, according to a preferred embodiment, the reaction chamber, at least partially, is made of a transparent material enabling optical detection in said reaction chamber.

The method of the present invention is preferably automated, e.g. carried out at least in part by an automatic analytical instrument, which is part of an analytical system.

The present invention is thus also directed to a system for carrying out heterogeneous chemical or biological reactions, the system comprising

- an analytical device, the analytical device comprising at least one liquid processing unit, the liquid processing unit comprising at least one reaction chamber and at least a first inlet channel in fluid communication with the reaction chamber,
- analyte capturing particles for capturing analytes contained in a liquid sample,
- at least a liquid flowing into the reaction chamber and generating a drag force Fd acting on the analyte capturing particles,
- an instrument for processing said analytical device, wherein said instrument comprises means for generating a counter-oriented force Fg acting on the analyte capturing particles.

According to one embodiment the analytical instrument comprises a rotatable supporting device and the analytical device is placed onto said rotatable supporting device so as to be able to rotate therewith.

The analytical instrument may further comprise a pipetting unit for supplying liquids to said reaction chamber via said at least one inlet channel.

The analytical instrument may further comprise a detection unit, e.g. an optical unit for detecting the result of the reactions.

The rotatable supporting device may be made e.g. of plastic or metal such as aluminium and may have a disk-like shape and can be rotated around a rotational axis driven by means of an actuator such as an electric motor.

The pipetting unit may comprise a reusable washable needle, e.g. a steel needle, or disposable pipette tips. Typically, the pipetting unit is operatively coupled to an automated positioning device for moving the pipette tip or needle with respect to the analytical device and, e.g., may be mounted to a transfer head that can be moved in two directions of travel in a plane, e.g., by means of guiding rails and a third direction of travel orthogonal to the plane, e.g., by means of a spindle drive.

The detection unit may comprise a light source, e.g. a xenon lamp, a laser, or LEDs, the optics, e.g. mirrors, lenses, optical filters, fiber optics, for guiding and filtering the light, one or more reference channels, a CCD sensor or the like. The detection unit is e.g. adapted for detecting analytes captured by the analyte capturing particles. For example, it may be embodied as a fluorescence detector for detecting fluorescence light emitted from fluorescence markers made to be bound to analytes captured by the analyte capturing particles.
The analytical system may further comprise a controller for controlling the automated analysis of samples according to a predetermined process operation plan which, e.g., may be embodied as programmable logic controller running a computer-readable program provided with instructions to perform operations in accordance with the process operation plan.
The analytical system may further comprise a reagent rack for receiving reagent containers, a sample rack for receiving sample containers, a washing station for washing e.g. the pipetting needle, a shaker or mixing paddle for suspending analyte capturing particles contained in liquid reagent containers, etc…

Supplying liquids to the reaction chamber via the at least one inlet channel is preferably carried out automatically by means of a pipetting unit as described with reference to the analytical instrument.

Detection of the signal generated by the signal generating analytes is preferably carried out by means of an optical detection unit as described with reference to the analytical system and may be based on photometric methods chosen from the group of absorbance measurement, turbidimetry, luminescence, bioluminescence, chemiluminescence, fluorescence, phosphorescence.

Since temperature may play an important role for chemical and biological reactions, the method may comprise the step of heating and/or cooling at least the reaction chamber, wherein heating and cooling generally mean providing heat to or subtracting heat from the reaction chamber. This comprises maintaining the liquid contained in the reaction chamber

at a constant temperature and/or subjecting the liquid to temperature changes, e.g. temperature gradients, repeated temperature cycles, etc…

Different ways of heating and cooling are possible. This may be achieved for instance by means of temperature regulating units, e.g. , Joule heating elements, infrared heating elements or Peltier elements, in contact with the reaction chamber, or by means of air streams. The all analytical device may be contained e.g. in a temperated housing of the system.

Other and further objects, features and advantages of the invention will appear from the following description and accompanying drawings, which illustrate preferred embodiments and serve to explain the principles of the invention more in detail.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is a partial perspective top view of an exemplary analytical device showing the elements of a possible liquid processing unit.

FIG. 2 shows schematically one embodiment of reaction chamber according to the flow-through variant.

FIG. 3 helps to understand the principle of confining the analyte capturing particles in a particle rearrangement zone.

FIG. 4 shows schematically other embodiments of reaction chambers.

FIG. 5 shows an embodiment comprising a dosing chamber.

FIG. 6 shows schematically an analytical system comprising an analytical device such as that shown in FIG. 1.

FIG. 7 shows schematically one embodiment of reaction chamber according to the shake-mode variant.

FIG. 8a and 8b help to understand the principle of fluidic movements caused by Euler forces according to the shake-mode variant.

DETAILED DESCRIPTION OF THE INVENTION

Figure 1 shows an analytical device 10 for heterogeneous chemical or biological reactions comprising a device body 11 with a disc-like shape. The device body 11 comprises a plurality of liquid processing units 20. The liquid processing unit 20 comprises a reaction chamber 21, for mixing at least one liquid with analyte capturing particles and two inlet channels, a first inlet 22, and a second inlet channel 23 in fluid communication with the reaction chamber 21 for delivering liquids to the reaction chamber 21.

The liquid processing unit further comprises a first inlet chamber 26 in fluid communication with the reaction chamber 21 via the first inlet channel 22 and a second inlet chamber 27 in fluid communication with the reaction chamber 21 via the second inlet channel 23. The liquid processing unit 20 comprises also an outlet channel 28 and one waste chamber 29 in fluid communication with the reaction chamber 21 via the outlet channel 28 for receiving liquids flowing out of the reaction chamber 21. A vent 30 in fluid communication with the waste chamber 29 allows air to escape as liquids flow into the reaction chamber 21 and eventually into the waste chamber 29.

The analyte capturing particles are introduced into the reaction chamber 21 via the second inlet channel 23 after pipetting a volume of liquid suspension into the second inlet chamber 27. Other liquids are introduced into the reaction chamber 21 via the first inlet channel 22 after pipetting respective volumes into the first inlet chamber 26.

The device body 10 is made of an injection-moulded polymer and is covered by a thin foil (not shown) sealing the liquid processing units 20. Access ports or holes are provided in correspondence of the inlet chambers 26, 27 and vent 30 in order to allow liquids to be introduced and air to escape.

The reaction chamber 21 is designed as a particle rearrangement zone 24, in which, due to an equilibrium of forces, the particles 12 are able to rearrange themselves, but from which the particles 12 are unable to escape, said forces comprising a drag force Fd generated by liquids flowing through the first inlet channel 22 and a counter-oriented force Fg. This principle is illustrated schematically in fig. 2 and 3. In this case, the counter-oriented force Fg is a centrifugal force, i.e. an acceleration force, generated by rotating the analytical device 10. The drag force Fd is dependent on centrifugal force as well as liquids flow into the reaction chamber 21 from the inlet chamber 26 during rotation of the analytical device 10. The average flow velocity of the liquids in the reaction chamber is however dependent also on the dimensions of the first inlet channel 22 and the geometry of the reaction chamber 21. Particularly, the average flow velocity is made to gradually decrease from the bottom (zone A in

fig. 3) to the top (zone C in fig. 3) of the reaction chamber 21. This is achieved e.g. by designing the reaction chamber 21 with a tapered shape, in this case with diverging walls in the flow direction, which is from A towards C. As the flow velocity decreases also the drag force Fd acting on the particles 12 decreases. Although, parallel walls could be used as well, having diverging walls allows to have larger particle rearrangement zones 24 and reduces the risks that particles 12 escape from the reaction chamber 21 via the outlet channel 28. While liquids may overflow out of the reaction chamber 21 via the outlet channel 28, the particles 12 remain, dynamically rearranging themselves, in the particle rearrangement zone 24 while liquids flow through the first inlet channel 22, and are compacted at the bottom of the reaction chamber 21 when liquids stop flowing through the first inlet channel 22. For limiting the maximum flow velocity and better controlling the equilibrium of forces, a narrow and long first inlet channel 22 or the presence of a flow resistor 31 is used. This has also the effect to prolong the reaction time while the slow supply of fresh liquids, e.g. reagents, makes reactions more efficient, i.e. by shifting the reaction equilibrium towards product formation. The counter-oriented force Fg acting on the particles 12 determines how far the particles 12 move into direction of zone C, which is how large the particle rearrangement zone 24 is. If the force Fg increases the particle rearrangement zone 24 will be small and located at the bottom of the reaction chamber 21. If the force Fg decreases, the particles 12 will be more distributed, i.e. the particles rearrangement zone 24 will occupy most of or all the reaction chamber 21. Thus varying the geometry of the reaction chamber 21 and the first inlet channel 22 and adjusting the rotational speed of the analytical device 10, the point (zone B in fig. 3) at which the forces Fd and Fg acting on the particles 12 in the reaction chamber 21 reach an equilibrium varies and so does the size and location of the particle rearrangement zone 24. In zone C Fg overcomes Fd and any particle 12 entering that zone tends to return towards B. In zone A Fd overcome Fg and any particle 12 in that zone tends to move towards B. As a result the particles 12 rearrange themselves and are trapped in the particle rearrangement zone 24 until both forces Fd and Fg are maintained.

The reaction chamber 21 may have different shapes and opening-angles that influence the flow-velocity profile in a different way. Beside one diverging shape, multiple diffuser-like compartments, organized in parallel or in series may be implemented. The ideal geometry creates a decreasing velocity gradient in flow-direction only and a constant uniform velocity in the orthogonal direction. However in practice a parabolic flow-profile is very likely to occur. The suspended particles 12 might get collected, e.g. accumulate or form aggregates, in low-velocity zones, such as in correspondence of stepped or sharply diverging walls. This unwanted effect may be promoted by rough or electrostatically charged surfaces. Therefore rather steep and fluent contours are preferred, eventually with structures promoting a uni-

form flow profile. The geometry may be designed such that the suspended material is collected at the end of a reaction in a dedicated area 32 of the reaction chamber 21 for e.g. detection purposes as shown in fig. 2.

In fig. 4 other examples of reaction chamber 21 designed to function at least in part as particle rearrangement zone 24 are schematically shown. In fig. 4a a symmetrically diverging reaction chamber 21 is shown. In fig. 4b an asymmetrically diverging reaction chamber 21 is shown. In fig. 4c the reaction chamber 21 comprises a structure 33 to split the flow in many parallel streams, that is like having multiple parallel rearrangement zones. This structure provides a more uniform flow profile by flattening the flow profile and causes the particles 12 to be more distributed in the centre of the reaction chamber 21 and less on the outer walls. In fig. 4d diffuser-like compartments 34 in series where each compartment 34 traps a fraction, e.g. size dependent, or a part of the particle 12 is shown. Fig. 4e is just a variant of fig. 4a wherein the position of the inlet channel 22 varies and is similar to the embodiment shown in fig. 1 and 2.

In other embodiments, filters (not shown) could also be used, although one object of the invention is to maintain the analytical device as simple as possible and the cost of manufacturing low.

Fig. 5 shows the structure of an exemplary dosing chamber 35 in fluid communication with the reaction chamber 21 via the first inlet channel 22, for delivering a defined volume of liquid, e.g. sample, to the reaction chamber 21. The volume defined by the liquid dosing chambers 35 is about 200 nanoliters. The liquid dosing chamber 35 has an elongated shape and three microchannels connected to it: a third liquid inlet channel 36 in fluid communication with the first inlet chamber 26 (not shown in fig. 5) allowing liquid to fill the liquid dosing chamber; a liquid decanting channel 37, defining where the liquid dosing chamber 35 starts and the third liquid inlet channel 36 ends, and allowing excess liquid to be guided to a waste chamber (not shown); the first inlet channel 22 in fluid communication with the reaction chamber 21. At the interface between the dosing chamber 35 and the sample dosing chamber 22 a geometric valve 38 is located. At this position the sample flow will temporarily stop and any excess of sample will be deviated to the decanting channel 37. Afterwards the driving force, in this case the centrifugal force, is increased above that required to break the energy barrier of the valve 38 and 200 nanoliters of liquid sample will be transferred to the reaction chamber 21 via the first inlet channel 22.

Fig. 6 shows schematically an analytical system 40 for heterogeneous chemical or biological reactions comprising an instrument, the instrument comprising a rotatable supporting device 41. An analytical device 10 as that of fig. 1 is removably fixed to said supporting device 41 so as to rotate therewith. The instrument further comprises a needle 44, part of a pipetting unit (not shown), for supplying liquids to the reaction chambers 21 via the inlet channels 22,23, an optical unit 45 for detecting the result of the reactions, a reagent rack 46 for receiving reagent containers, a sample rack 47 for receiving sample containers, a washing unit 48 for washing the needle 44 of the pipetting unit.

FIG. 7 shows schematically one embodiment of reaction chamber according the shake-mode variant. The reaction chamber 71 is divided into two sub-chambers 71a and 71b which are connected by a connecting section 71c which is located on the side of the reagent chamber 71 which is most distant from the rotational axis. This partition of the reaction chamber 71 into two sub-chambers 71a and 71b is caused by a solid and raised structure 74. This solid structure provokes that the flow of liquid between the two sub-chambers 71a and 71b is directed to the connecting section 71c. In addition to the connecting section 71c on the side of the reaction chamber 71 which is more distant from the rotational axis, an additional connection 75 between the sub-chambers 71a and 71b is given on the side of the reaction chamber 71 which is closer the rotational axis. This additional connection 75 can be used as a volume compensation area when fluids are transported between the two sub-chambers 71a and 71b in the shake-mode. In the embodiment of figure 7, the rotational axis is located above the shown fluidic structure.

A first inlet channel 72 and a second inlet channel 73 are in fluid communication with the reaction chamber 71. These inlet channels 72 and 73 can be used for the application of liquids into the reaction chamber, e.g. for the application of reagent liquids or washing liquids. Also an outlet channel 78 is in fluid communication with the reaction chamber 71 for receiving liquids flowing out of the reaction chamber 71. This outlet channel is preferably in fluid communication with a waste chamber (not shown) which receives and stores all liquids used in the reagent chamber 71. In a preferred embodiment, the outlet channel 78 is designed as a microfluidic siphon (see figure 7).

In figure 7, a vent 80 in fluid communication with the reaction chamber 71 allows air to escape as liquids flow into the reaction chamber 71.

Recesses 76 are also shown in figure 7. These recesses 76 are preferably depressions in the bottom wall of the reaction chamber 71 and can be advantageously used for a defined ap-

plication of reagents and also particles into the reaction chamber 71 during manufacturing of the analytical device. Preferably, liquid reagents or particle suspensions are provided and dried into these recesses 76 during manufacturing of the analytical device. During use of the analytical device, the reagents or particles are solubilised or resuspended by contact with liquids (e.g. the sample or buffer solutions) which saves an additional reagent application step during use of the analytical device.

The analyte capturing particles are either introduced into the reaction chamber 71 via the inlet channel 72, 73 or be already present, preferably dried into the recesses 76 and resuspended by a liquid entering the reaction chamber 71 via the inlet channel 72, 73. In alternative embodiments, the analyte capturing particles can be provided at different sites within the reaction 71, e.g. in the connecting section 71c. Preferably, the particles have a higher density than the surrounding liquid. As a result of this higher density, the particles are agglomerated at the section f the reaction chamber 71 which is most distant from the rotational axis when applying centrifugal forces. This is the connecting section 71c. Therefore, in a preferred embodiment this connecting section 71c is also used as the detection area, because within this area the particles can be agglomerated in a high packing density which is very beneficial for high signal intensity and sensitivity. For these reasons, the connection section 71c is made of transparent material or comprises at least an optical window which allows a signal detection by optical methods.

The preferred designs of other embodiments, e.g. the geometric features shown in and described in context with figures 1 to 5 can be used analogously for the shake-mode variant according to figure 7.

Figures 8a and 8b show schematically two different states respectively occurring in a reaction chamber very similar to the reaction chamber shown in figure 7 during use and application of a shake-mode acceleration profile. The darker area in the lower part of the structure represents the arrangement of particles located within this connecting section of the reaction chamber and the lighter area above the darker particle area represents the liquid which is “pumped” through the particle arrangement in the shake-mode. In addition to the light area, liquid is of course also present within the dark area, filling the spaces between the particles. During the shake-mode, Euler forces are appearing which cause an effective flow of liquid from one sub-chamber to the other sub-chamber. These alternating effective liquid flows through the particle arrangement result in an improved binding efficiency for analytes onto the binding surfaces of the particles.

In the following an example of method for carrying out heterogeneous chemical or biological reactions according to the invention is given.

The method comprises the steps of:

a) placing an analytical device 10 such as that of fig. 1 in the instrument of system 40,

b) supplying to the reaction chamber 21 via the second inlet channel 23 analyte capturing particles 12, or providing analyte capturing particles in the reaction chamber 71, preferably by a analyte capturing particles pre-loading step during manufacturing of the analytical device,

c) supplying to the reaction chamber 21, 71 via the first inlet channel 22, 72 a liquid sample containing at least an analyte of interest,

d) confining by an equilibrium of forces the analyte capturing particles 12 in a particle rearrangement zone 24 comprised in the reaction chamber 21, 71, said forces comprising a drag force Fd, generated by the liquid sample flowing through the first inlet channel 22 or induced by Euler forces in a shake-mode, and a counter-oriented centrifugal force Fg generated by rotating the analytical device 10,

e) capturing analytes present in the liquid sample with the particles 12 in the particle rearrangement zone 24 while the liquid sample is flowing.

In the following another more detailed example of method for carrying out heterogeneous chemical or biological reactions according to the invention is given. The method comprises the steps of

a) placing an analytical device 10 such as that of fig. 1 in the instrument of system 40,

b) supplying by the pipetting unit a liquid suspension comprising analyte capturing particles 12 to the second inlet chambers 27,

c) transferring by rotating the analytical device 10 said first liquid reagent to the reaction chambers 21 via the second inlet channels 23,

d) supplying by a pipetting unit liquid samples to the first inlet chambers 26,

e) transferring by rotating the analytical device 10 the liquid samples to the reaction chambers 21 via the first inlet channels 22,

f) capturing analytes present in the liquid samples by means of the analyte capturing particles 12,

g) supplying by a pipetting unit to the first inlet chambers 26 a first liquid reagent comprising signal generating conjugates for binding to analytes captured by the analyte capturing particles 12,

h) transferring by rotating the analytical device 10 said first reagent to the reaction chambers 21 via the first inlet channels 22,

i) supplying by a pipetting unit to the first inlet chambers 26 a wash buffer for washing out bound-free signal generating conjugates,

j) transferring by rotating the analytical device 10 said wash buffer to the reaction chambers 21 via the first inlet channels 22,

k) repeating steps i) and j) as necessary for sufficient washing to let previous liquids contained in the reaction chambers and excess of washing buffer flow out of the reaction chambers 21 into the waste chambers 29 via the outlet channels 28,

l)confining by an equilibrium of forces at least temporarily during steps c), e), f), h), j) the analyte capturing particles 12 in the particle rearrangement zones 24 comprised in the reaction chamber 21, said forces comprising a drag force Fd generated by liquids flowing through the inlet channels 22 and a counter-oriented centrifugal force Fg,

m) collecting the particles 12 carrying the conjugate-bound analytes in the areas 32 by centrifugal force Fg only,

n) detecting the signal generated by the conjugate-bound analytes captured by the particles 12.

Obviously many modifications and variations of the present invention are possible in light of the above description. It is therefore to be understood, that within the scope of appended claims, the invention may be practiced otherwise than as specifically devised.

CLAIMS

1. Method for carrying out heterogeneous chemical or biological reactions comprising the steps of:

a) providing an analytical device (10) comprising at least one liquid processing unit (20), the liquid processing unit (20) comprising at least one reaction chamber (21, 71), and at least a first inlet channel (22, 72) in fluid communication with the reaction chamber (21,71),

b) supplying to the reaction chamber (21, 71) via the first inlet channel (22, 72) or a second inlet channel (23, 73) analyte capturing particles 12, or providing analyte capturing particles (12) in the reaction chamber (21, 71), preferably by a analyte capturing particles pre-loading step during manufacturing of the analytical device,

c) supplying to the reaction chamber (21, 71) via the first inlet channel (22, 72) or the second inlet channel (23, 73) a liquid sample containing at least an analyte of interest,

d) confining by an equilibrium of forces the analyte capturing particles (12) in a particle rearrangement zone (24) comprised in the reaction chamber (21, 71), said forces comprising a drag force Fd, generated by liquids flowing through the first inlet channel (22, 72) or generated by liquids flowing induced by Euler forces within the reaction chamber (21, 71), and a counter-oriented force Fg,

e) capturing analytes present in the liquid sample with the particles (12) in the particle rearrangement zone (24).

2. The method according to claim 1 further comprising the steps of:

f) supplying to the reaction chamber (21, 71) via the first inlet channel (22, 72) a first liquid reagent comprising signal generating conjugates for binding to analytes captured by the analyte capturing particles (12) or providing a reagent comprising signal generating conjugates for binding to analytes captured by the analyte capturing particles in the reaction chamber (21, 71), preferably by a reagent pre-loading step during manufacturing of the analytical device,

g) supplying to the reaction chamber (21, 71) via the first inlet channel (22, 72) a second liquid reagent for washing unbound signal generating conjugates out of the particle rearrangement zone (24),

h) detecting a signal generated by the conjugate-bound analytes captured by the particles (12).

3. The method according to claim 1 or 2 wherein the counter-oriented force Fg is induced by a field chosen from the group of gravitational field, magnetic field, electromagnetic field,

electrostatic field, acceleration field comprising centrifugal, translational and rotational acceleration fields.

4. The method according to any of the preceding claims wherein said reaction chamber (21) has a tapered shape whereby the downstream directional flow velocity of liquids flowing in the reaction chamber (21) via said first inlet channel (22) is reduced.

5. The method according to any of the preceding claims wherein the counter-oriented force Fg is induced by a centrifugal field and wherein the drag force Fd is induced by an Euler force which is generated by acceleration of the analytical device (10).

6. The method according to any of the preceding claims comprising the step of splitting liquids flowing through the reaction chamber (21, 71) in multiple streams.

7. The method according to any of the claims 1 to 4 comprising the step of dividing the reaction chamber (21, 71) in serial diffuser-like compartments (34).

8. The method according to any of the preceding claims comprising the step of dosing defined volumes of liquids to be delivered to the reaction chamber (21, 71) via at least one liquid dosing chamber (35) in fluid communication with the reaction chamber (21, 71) via at least said first inlet channel (22, 72).

9. The method according to any of the preceding claims comprising the step of inducing liquids to flow out of the reaction chamber (21, 71) into a waste chamber (29) in fluid communication with the reaction chamber (21, 71) via an outlet channel (28, 78).

10. The method according to any of the preceding claims wherein the first channel (22, 72) comprises a flow resistor (31) for controlling the flow velocity of liquids .

11. The method according to any of the preceding claims comprising the step of rotating the analytical device (10).

12. The method according to any of the preceding claims comprising the step of heating or cooling at least the reaction chamber (21, 71).

13. The method according to any of the preceding claims comprising the step of agglomerating the particles (12) by centrifugal forces in a detection area within the reaction chamber (21, 71) which is most distant from the rotational axis used to generate the centrifugal forces.

14. The method according to any of the claims 2 to 13 wherein detecting the signal generated by the conjugate-bound analytes captured by the particles (12) is based on photometric methods chosen from the group of absorbance measurement, turbidimetry, luminescence, bioluminescence, chemiluminescence, fluorescence, phosphorescence.

15. System for carrying out heterogeneous chemical or biological reactions comprising
- an analytical device (10), the analytical device (10) comprising at least one liquid processing unit (20), the liquid processing unit (20) comprising at least one reaction chamber (21, 71) and at least a first inlet channel (22, 72) in fluid communication with the reaction chamber (21, 71),
- analyte capturing particles (12) for capturing analytes contained in a liquid sample,
- at least a liquid flowing into the reaction chamber (21, 71) and generating a drag force Fd acting on the analyte capturing particles (12),
- an instrument for processing said analytical device (10), wherein said instrument comprises means for generating a counter-oriented force Fg acting on the analyte capturing particles (12).

16. The system according to claim 15 wherein the counter-oriented force Fg is induced by a centrifugal field and said reaction chamber (71) is divided into two sub-chambers (71a, 71b) which are connected by a connecting section (71c) which is located on the side of the reagent chamber (71) which is most distant from the rotational axis used to generate the centrifugal field.

17. The system according to one of the claims 15 or 16 wherein the instrument further comprises a pipetting unit for supplying liquids to the analytical device (10).

18. The system according to one of the claims 15 to 17 wherein the instrument comprises an optical unit (45) for detecting the result of the reactions.

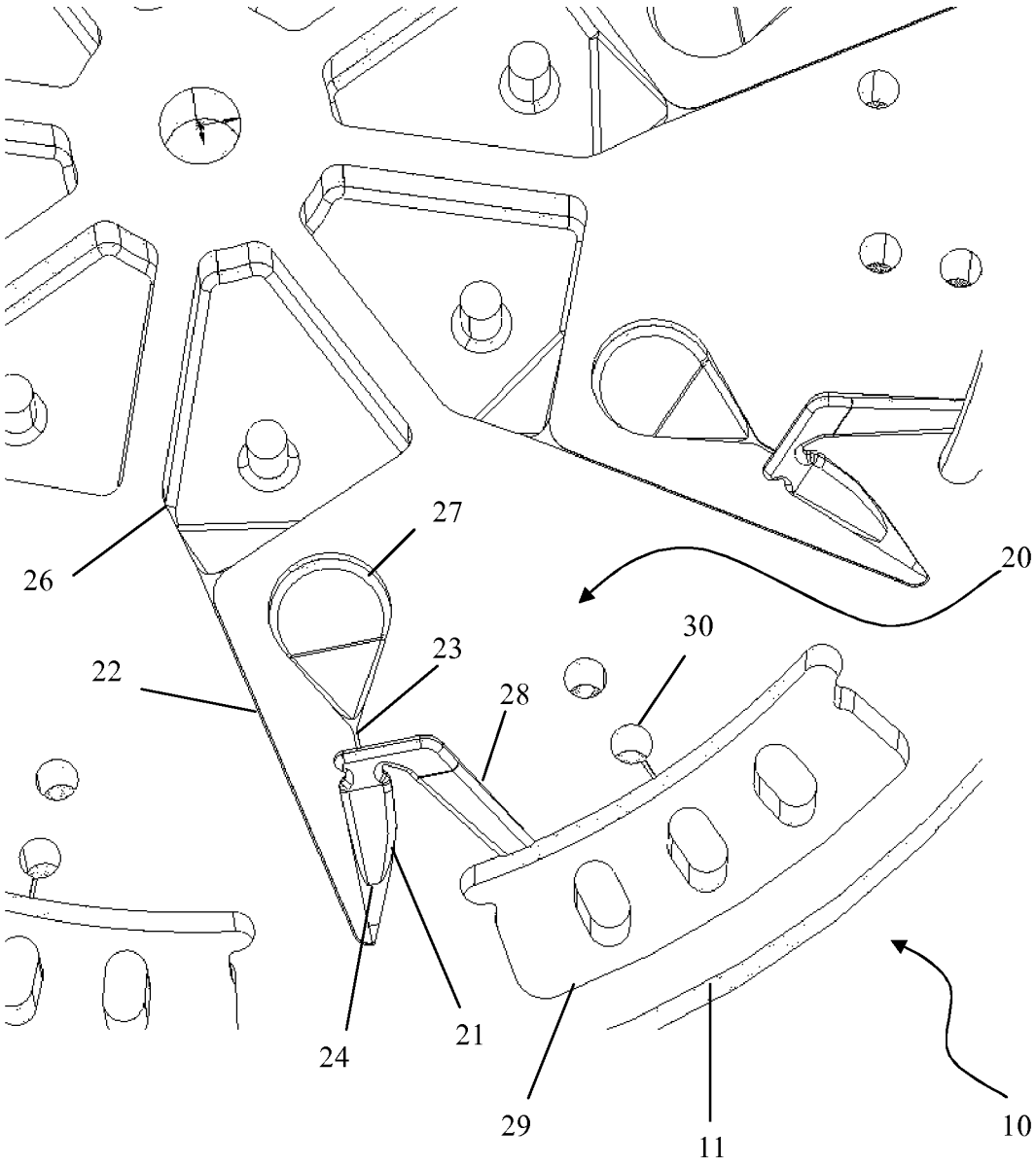

Fig. 1

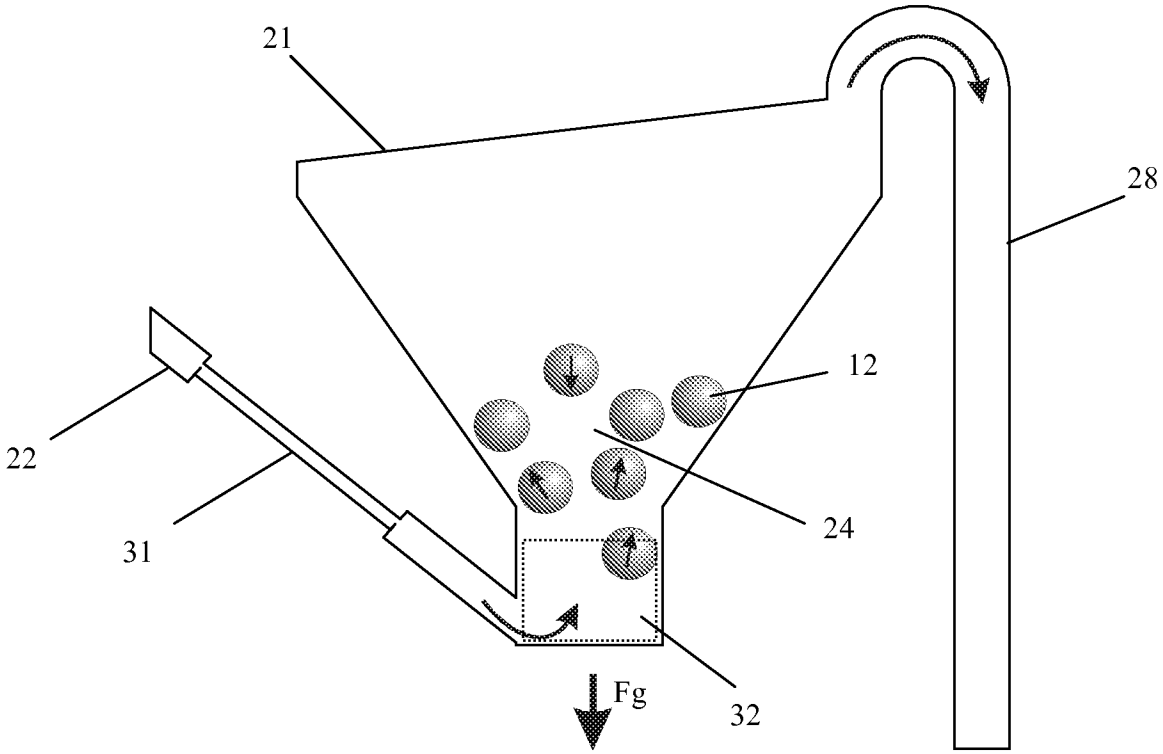

Fig. 2

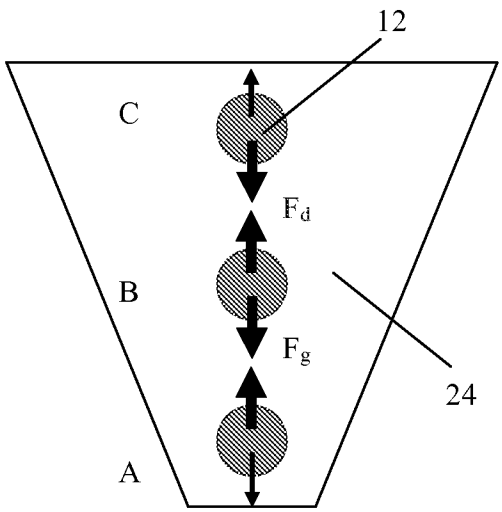

Fig. 3

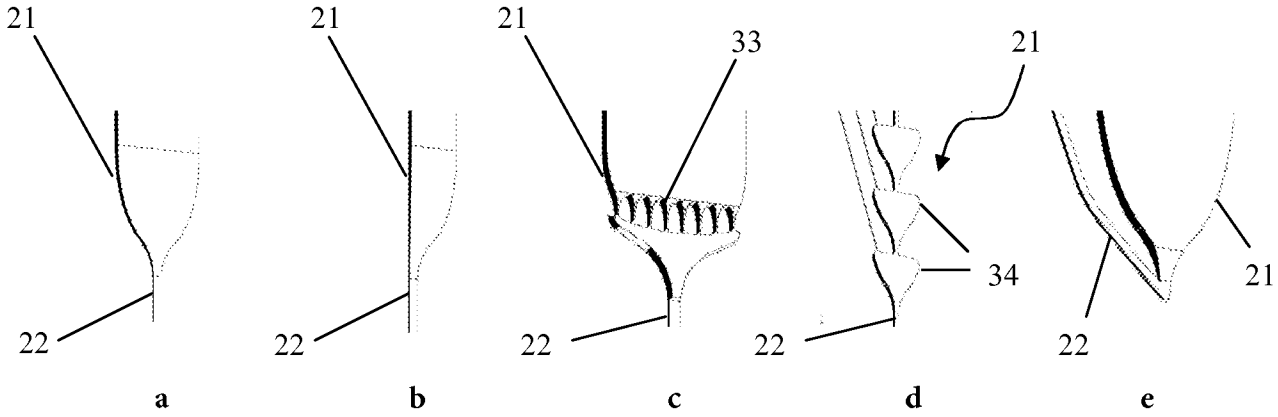

Fig. 4

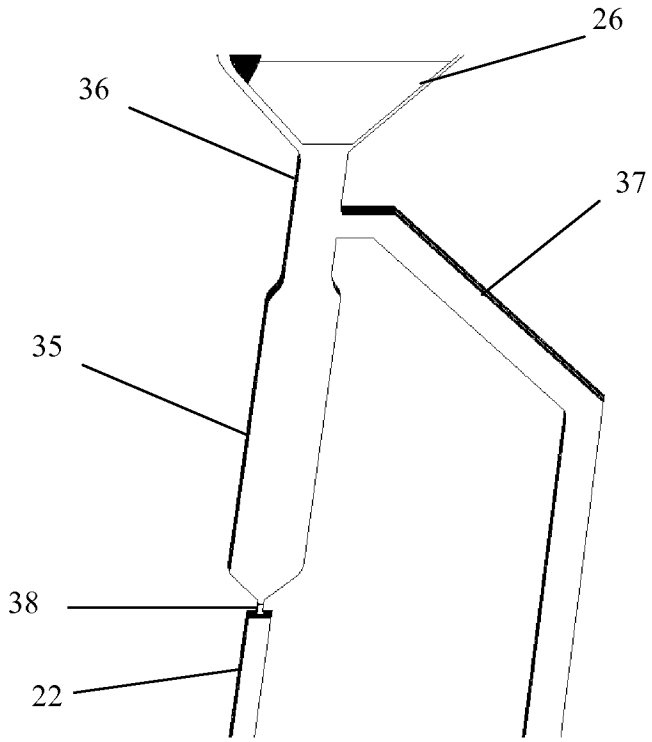

Fig. 5

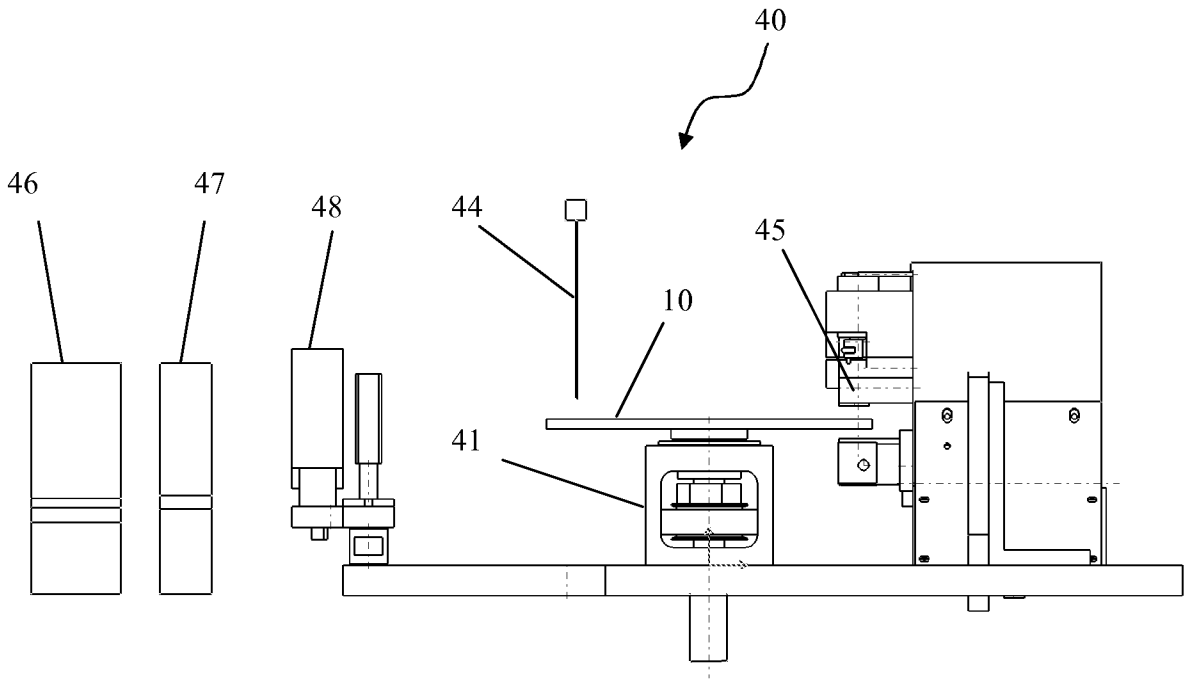

Fig. 6

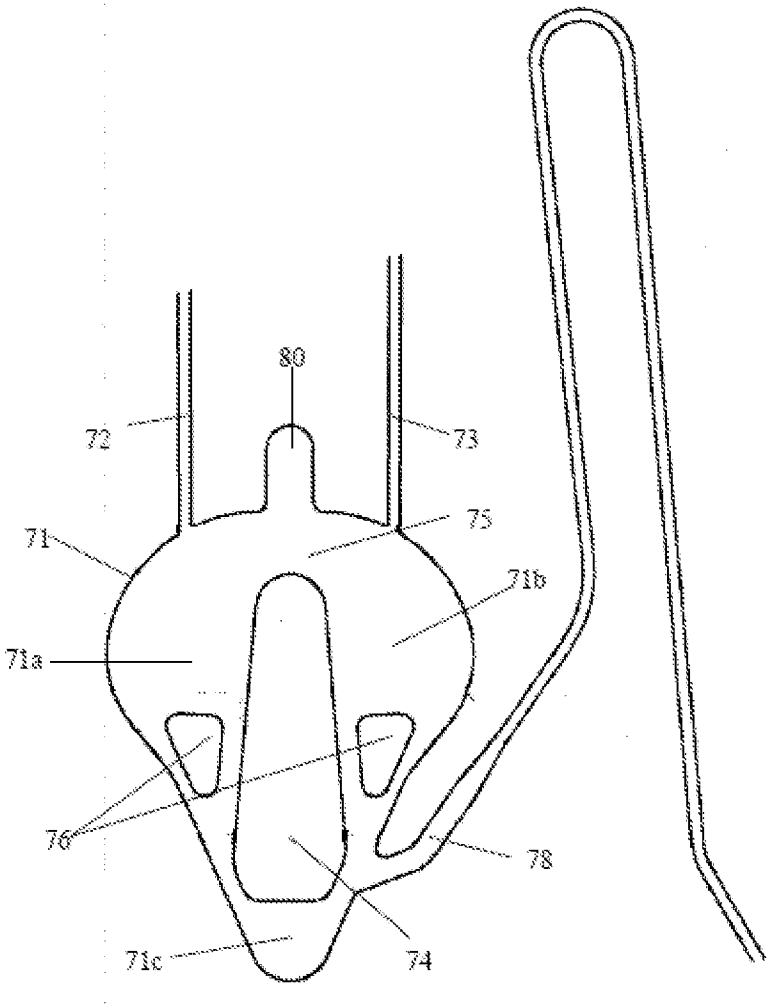

Fig. 7

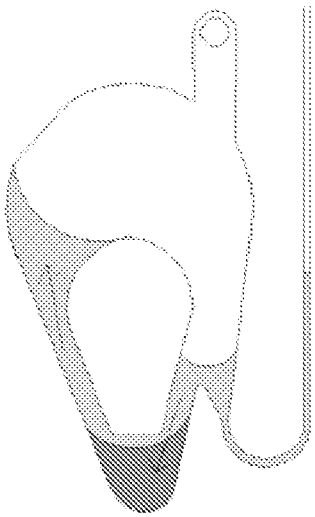

Fig. 8a

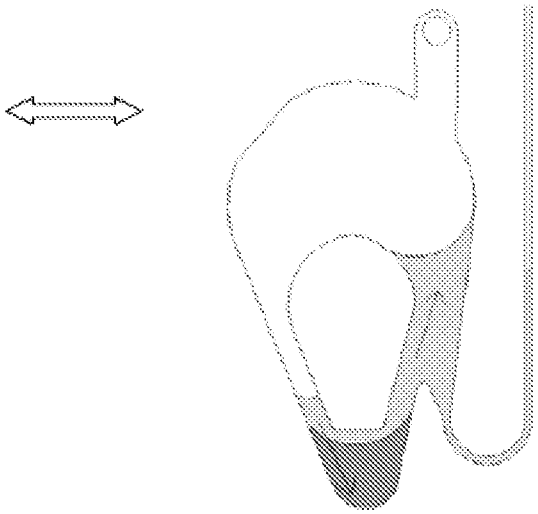

Fig. 8b

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2010/063477

A. CLASSIFICATION OF SUBJECT MATTER
INV. G01N33/543 B01L3/00 G01N35/00 B04B5/04
ADD.

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)
G01N B01L B04B

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 practical, search terms used)
EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2007/050619 A1 (HEWLETT PACKARD DEVELOPMENT COM; TYVOLL D; JOHNSON B) 3 May 2007 (2007-05-03)	15,17,18
A	the whole document, in particular p. 8, l. 15-27; p. 9, l. 4-22; p. 10, l. 23-26; figures 3, 4A, 4B	1-14,16
X	WO 98/50163 A1 (COBE LAB) 12 November 1998 (1998-11-12)	15,17,18
A	the whole document, in particular p. 17, l. 19-24; p. 33, l. 35 to p. 34, l. 6; figures 1 and 3	1-14,16

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

[X] See patent family annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
13 October 2010	27/10/2010

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Weber, Peter

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2010/063477

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

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X	EP 1 901 054 A2 (SAMSUNG ELECTRONICS CO LTD) 19 March 2008 (2008-03-19)	15,17,18
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A	WO 2007/106013 A1 (GYROS PATENT AB; ANDERSSON P; JESSON G) 20 September 2007 (2007-09-20) the whole document, in particular p. 5, l. 18-19; claim 1, 15, 16	1-18
A	LUTZ S ET AL: "UNIDIRECTIONAL SHAKE-MODE FOR MIXING HIGHLY WETTING FLUIDS ON CENTRIFUGAL PLATFORMS" The proceedings of mTAS 2008 conference 12 October 2008 (2008-10-12), pages 748-750, XP002604717 ISBN: 978-0-9798064-1-4 Retrieved from the Internet: URL:http://www.rsc.org/binaries/LOC/2008/PDFs/Papers/252_0388.pdf [retrieved on 2010-10-12] the whole document	1-18

INTERNATIONAL SEARCH REPORT

Information on patent family members

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