

to add the liquid to particles, retrieving the liquid from particles requires a higher level of hands-on skills. One needs to carefully position the liquid handling device, so that maximal amount of waste liquid could be removed without disturbing the particles. If too much liquid is left in the residue, it would lead to a high level of carry-over contamination. If particles are removed with the waste liquid by accident, it would result in a low yield or a low detection sensitivity. Accordingly the performance of particle-based systems is often compromised by the carry-over contamination caused by the residual reagents during the liquid-exchange process.

There is therefore a need to efficiently separate the particles of particle-based assays from the liquids used in various assay steps. A device that is able to effectively remove the waste liquid, thereby achieving a more efficient liquid exchange, as compared to the conventional process, and minimizing the carry-over contamination is therefore desired in the art.

Summary

The disclosure relates to a device for liquid removal in particle-based assay systems comprising (a) a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber (b) a porous or sieve-like membrane, attached to the chamber covering the at least one opening for removing the liquid, to prevent the flowing of a liquid out of the chamber when not in contact with an absorbent pad, wherein the membrane has pores or holes with a diameter that is smaller than the particle diameter of the particles used in the assay; and (c) a membrane-detachable liquid absorbent pad that can be attached to the outer side of the membrane not facing the inner chamber.

According to a first aspect, there is provided a device for liquid removal in particle-based assay systems comprising (a) a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber; (b) a porous or sieve-like membrane, attached to the chamber covering the at least one opening for removing the liquid, to prevent the flowing of a liquid out of the chamber when not in contact with an absorbent pad; wherein the membrane has pores or holes with a diameter that is smaller than the particle diameter of the particles used in the assay; and (c) a liquid absorbent pad that can be attached to and detached from the outer side of the membrane not facing the inner chamber.

Advantageously, the device according to the disclosure can be used to effectively remove the waste liquid from particles. The capillary-driven liquid-exchange process by the absorbent pad is able to efficiently separate (“wipe off”) the waste liquid from the particles, thereby reducing any carry-over contamination. The absorbent pad may provide a passive pumping mechanism for simple fluidic handling. In contrast, the conventional liquid exchange by centrifugal or magnetic force would leave a large amount of liquid behind on the particle surface as a result of surface tension, leading to a high level of contamination.

The removal with the absorbent pad comprised in the device allows reactions to incubate for a predefined duration in the fluidic handling chamber, while the absorbent pad is detached from the membrane. It removes the waste liquid when the absorbent pad is brought into contact with the membrane. It further allows for passive fluidic pumping, which eliminates the use of bulky and expensive external fluidic control systems.

The disclosure advantageously provides a capillary-driven vertical flow platform for liquid exchange in particle-based systems. The platform, referred to by the name "sieve-through", consists of a reaction unit and an absorbent pad (Fig. 1). The key feature of the sieve-through platform is the porous membrane that forms the bottom of the reaction chamber. Despite the fact that it is porous, the membrane is able to contain the liquid in the reaction unit due to surface tension. However, when the membrane is brought into contact with the absorbent pad, the capillary force provided by the absorbent pad

would pull the liquid out of the reaction unit through the pores. The pores of the membrane are smaller than the diameter of the particles used in the assay. As a result, particles would be sieved out and retained on the membrane during the removal of the waste liquid through the pores, hence the name "sieve-through".

According to a second aspect of the disclosure there is provided an array of devices according to the first aspect of the invention wherein the membranes of all or several devices can be contacted by a single absorbent pad or multiple sets of absorbent pads either simultaneously or at staggered sequence.

Advantageously, the array provides an example of the disclosure wherein this example has the potential for use in high-throughput analysis by presenting a sieve-array, which allows concurrent analysis of multiple samples in parallel. The sieve-through array may significantly improve the performance of particle-based systems.

According to a third aspect of the disclosure, there is also provided a preparative or quantitative assay method, comprising the steps of (a) providing a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber which is covered by a porous or sieve-like membrane; (b) filling particles and at least one liquid into the chamber together with a preparation of a sample material and optional reagents and/or other materials into the chamber; (c) performing a reaction in the chamber wherein the particles react or interact with a preparation of a sample material without any substantial flow of liquid through the membrane; (d) removing the reaction liquids and optionally dissolved by-products via the porous or sieve-like membrane by causing a flow through the membrane after contacting the membrane with an absorbent pad at the outer side of the membrane not facing the inner chamber; (e) optionally adding a liquid to the chamber; and (f) detecting or collecting the sample material that has reacted with the particle.

The method makes use of the fluidic handling device according to the first aspect of the disclosure. Advantageously, the inventive method can be used to handle the liquid exchange in particle-based enzyme-linked immunosorbent assay (ELISA). The inventive method for liquid removal by "sieving" effectively allows for a removal of the waste liquid during ELISA, resulting in a low background. The method can be used in a broad variety of assays with liquid removal steps and is not limited to quantitative assays, but can also be used for preparatory assays. One example therefore relates to a method wherein the particles interact with the sample material to form sample materials which are tagged by the particle by adherence or chemical binding. Advantageously, these tagged materials can be easily separated from the reaction solution and washed before further use.

According to a fourth aspect of the disclosure, there is provided a preparative or quantitative assay method, comprising the steps of (a) providing a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber which is covered by a porous or sieve-like membrane; (b) filling sample material in particle form or immobilized on particles in a liquid preparation and optional reagents or fillers into the chamber; (c) removing the liquids and optionally dissolved substances via the porous or sieve-like membrane by causing a flow through the membrane after

contacting the membrane with an absorbent pad at the outer side of the membrane not facing the inner chamber; (d) optionally adding a liquid to the chamber; and (e) detecting or collecting the sample material in particle form or immobilized on the particles. According to this method, sample materials that are already in particle form or tagged to particles can be separated and/or washed in a simple way. As such the method even allows analyzing “natural particles” such as cells. Advantageously, liquid removal or exchange is very simple.

Definitions

The following words and terms used herein shall have the meaning indicated:

Those skilled in the art will appreciate that the invention described herein is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variations and modifications. The invention also includes all of the steps, features, compositions and compounds referred to or indicated in this specification, individually or collectively, and any and all combinations or any two or more of said steps or features.

As used herein, the term “assay” refers to an investigative (analytic) procedure in laboratory medicine, pharmacology, environmental biology and molecular biology for qualitatively assessing or quantitatively measuring the presence, amount, or functional activity of a target entity (the analyte). Assay is used in a broad sense in this description and includes preparatory assay methods which are directed to removing the analyte from a preparation to determine its amount or to use it further.

As used herein, the term “bioassay” refers to a procedure for determining the concentration, purity, and/or biological activity of a substance (e.g., vitamin, hormone, plant growth factor, antibiotic, enzyme) by measuring its effect on an organism, tissue, cell, enzyme or receptor preparation compared to a standard preparation.

As used herein, the term “ELISA” refers to analytic biochemistry assay that uses a solid-phase enzyme immunoassay (EIA) to detect the presence of a substance in a liquid sample or wet sample.

As used herein, the term “about”, in the context of concentrations of components of the formulations, typically means $\pm 5\%$ of the stated value, more typically $\pm 4\%$ of the stated value, more typically $\pm 3\%$ of the stated value, more typically, $\pm 2\%$ of the stated value, even more typically $\pm 1\%$ of the stated value, and even more typically $\pm 0.5\%$ of the stated value.

Unless specified otherwise, the terms “comprising” and “comprise”, and grammatical variants thereof, are intended to represent “open” or “inclusive” language such that they include recited elements but also permit inclusion of additional, not recited elements.

The word “substantially” does not exclude “completely” e.g. a composition which is “substantially not leaking” may be completely tight without any leakage. Where necessary, the word “substantially” may be omitted from the definition of the invention.

Claims

1. A device for liquid removal in particle-based assay systems comprising
 - a) a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber;
 - b) a porous or sieve-like membrane, attached to the chamber covering the at least one opening for removing the liquid, to prevent the flowing of a liquid out of the chamber when not in contact with an absorbent pad; wherein the membrane has pores or holes with a diameter that is smaller than the particle diameter of the particles used in the assay; and
 - c) a liquid absorbent pad that can be attached to and detached from the outer side of the membrane not facing the inner chamber.

2. The device according to claim 1 wherein a liquid can be removed from the chamber by vertical flow and the at least one opening for adding reagents is on top or at an upper section of the chamber and the at least one other opening for removing liquids from the chamber is at the bottom or at a lower section of the chamber, or wherein the flow of a liquid through the membrane is a passive flow after contact with the absorbent pad.

3. The device according to any of the preceding claims wherein the membrane becomes permeable for the liquids upon contact with the liquid absorbent pad, or wherein the membrane is a microporous membrane, or wherein the membrane prevents the liquid from flowing from the chamber with a hold-up time of more than 60 minutes, or wherein the membrane has pores or holes of a size of 0.05 to 50 μm .

4. The device according to any of the preceding claims wherein the chamber is a reaction chamber for performing a chemical or biochemical reaction in a liquid.

5. The device according to any of the preceding claims wherein the membrane is made from a non-absorbent polymer, or wherein the membrane is selected from polycarbonates, polyamides, modacrylic copolymers, styrene-acrylic acid copolymers, polysulfones, polyvinylidene fluoride, polyvinylfluoride, polychloroethers, thermoplastic polyethers, acetal polymers, polyacrylonitrile, polymethyl methacrylate, poly n-butyl methacrylate, polyurethanes, polyimides, polybenzimidazoles, polyvinyl acetate, aromatic and aliphatic polyethers, cellulose esters, epoxy resins, polyethylene, polypropylene, porous rubber, poly(ethylene oxides, polyvinylpyrrolidones, poly(vinyl alcohols), poly(sodium styrenesulfonate), polyvinylbenzyltrimethyl-ammonium chloride, poly(hydroxyethyl methacrylate), poly(isobutyl vinyl ether), polyisoprenes, polyalkenes, ethylene vinyl acetate copolymers, polyamides, and polyurethanes.

6. The device according to any of the preceding claims wherein the absorbent pad is made from a hydrophilic material.
7. The device according to any of the preceding claims wherein the particle based assay is preparative or analytical bioassay.
8. The device according to any of the preceding claims wherein the particle is optionally functionalized and is selected from silica particles, polymeric particles, magnetic or superparamagnetic particles, or wherein the particles are deposited on the membrane or on the chamber inner walls.
9. An array of devices according to any of the preceding claims wherein the membranes of all or several devices can be contacted by a single absorbent pad or multiple sets of absorbent pads either simultaneously or at staggered sequence.
10. An array according to claim 9 wherein the array consist of a well plate wherein the membrane is attached to the bottom of the well plate by adhesive, double-sided tape, polydimethylsiloxane (PDMS) or thermal bonding of the membrane to the plate bottom.
11. An array according to claim 9 or 10 wherein the membrane is a polycarbonate membrane with micrometer-size holes.
12. A preparative or quantitative assay method, comprising the steps of:
 - a) providing a chamber with at least one opening for adding particles, liquids and optionally other materials;
and with at least one other opening for removing liquids from the chamber which is covered by a porous or sieve-like membrane;
 - b) filling particles and at least one liquid into the chamber together with a preparation of a sample material and optional reagents and/or other materials into the chamber;
 - c) performing a reaction in the chamber wherein the particles react or interact with a preparation of a sample material without any substantial flow of liquid through the membrane;
 - d) removing the reaction liquids and optionally dissolved by-products via the porous or sieve-like membrane by causing a flow through the membrane after contacting the membrane with an absorbent pad at the outer side of the membrane not facing the inner chamber;
 - e) optionally adding a liquid to the chamber; and
 - f) detecting or collecting the sample material that has reacted with the particle.
13. The method according to claim 12 wherein the chamber is filled with a washing liquid after step d) and thereafter this washing liquid is removed through the membrane by further contacting the membrane the absorbent pad before performing to step e) or f).
14. The method according to claim 12 wherein the particles are polymeric particles functionalized with a labelled capture antibody and the preparation of the sample material comprises a enzyme-labeled detector antibody, or wherein the particles interact with the sample

material to form sample materials which are tagged by the particle by adherence or chemical binding.

15. The method according to claim 12 wherein the preparation comprises DNA and the particle is a magnetic particle, or wherein the preparation comprises mRNA and the particle is a poly(T) conjugated magnetic particle.

16. A method according to claim 12 wherein the particles are first filled into the chamber in step b) together with a liquid, the liquid is removed and the particle deposited on the membrane and or/chamber walls before the sample material together with at least one liquid is filled in the chamber.

17. A method according to claim 12 wherein the sample material is tagged to a particle during step c).

18. A preparative or quantitative assay method, comprising the steps of:

- a) providing a chamber with at least one opening for adding particles, liquids and optionally other materials and with at least one other opening for removing liquids from the chamber which is covered by a porous or sieve-like membrane;
- b) filling sample material in particle form or immobilized on particles in a liquid preparation and optional reagents or fillers into the chamber;
- c) removing the liquids and optionally dissolved substances via the porous or sieve-like membrane by causing a flow through the membrane after contacting the membrane with an absorbent pad at the outer side of the membrane not facing the inner chamber;
- d) optionally adding a liquid to the chamber; and
- e) detecting or collecting the sample material in particle form or immobilized on the particles.

19. The method according to claim 18 wherein the chamber is filled with a washing liquid after step d) and thereafter this washing liquid is removed through the membrane by further contacting the membrane the absorbent pad before performing to step d) or e).

20. The method according to claim 18 wherein the sample material immobilized on particles comprises DNA or RNA and the particles are selected from silica beads or silica-coated magnetic beads, or wherein the sample material immobilized on particles comprises mRNA and the particles are selected from oligo(dT) magnetic beads.