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PROVISIONAL APPLICATION FOR PATENT COVER SHEET

This is a request for filing a PROVISIONAL APPLICATION FOR PATENT under 37 CFR § 1.53(c)

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TITLE	
DESTABILIZING EMULSIONS ON MICROFLUIDIC DEVICES	
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Respectfully submitted,

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DESTABILIZING EMULSIONS ON MICROFLUIDIC DEVICES

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FIELD OF THE INVENTION

The invention described herein pertains to the formation of aqueous emulsions on microfluidic devices and methods of use.

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BACKGROUND

Microfluidic devices are essentially miniaturized chemistry laboratories, featuring a series of micrometer-sized channels etched into a chip in which nanoliter-sized aqueous emulsions can be formed and analyzed. Such analyses can provide a wealth of information for biomedical and analytical chemistry studies. Because of their incredibly small sample sizes — thousands of times smaller in volume than a typical droplet — microfluidic “labs on a chip” are highly prized for providing rapid analysis at relatively low costs.

High throughput molecular screening (HTS) is the automated, rapid testing of thousands of distinct small molecules or probes in cellular models of biological mechanisms or disease, or in biochemical or pharmacological assays. Active compounds identified through HTS can provide powerful research tools to elucidate biological processes through chemical genetic approaches, or can form the basis of therapeutics or imaging agent development programs. HTS has experienced revolutionary changes in technology since the advent of molecular biology and combinatorial chemistry, and the incorporation of modern information management systems. Current HTS instrumentation allows screening of hundreds of thousands of compounds in a single day at a rate orders of magnitude greater than was possible a decade ago.

Enzymes can be analyzed and/or sorted by the extent to which they catalyze chemical reaction of a substrate (conversely, substrate can be analyzed and/or sorted by the level of chemical reactivity catalyzed by an enzyme). Cells can be sorted according to whether they contain or produce a particular protein, by using an optical detector to examine each cell for an optical indication of the presence or amount of that protein. The

protein may itself be detectable, for example by a characteristic fluorescence, or it may be labeled or associated with a reporter that produces a detectable signal when the desired protein is present, or is present in at least a threshold amount.

5 After the aqueous emulsions have been sorted and collected, the components of the emulsions need to be collected. Thus, what is needed is a simple method to permit recovery of the aqueous phase components (cells, enzymes, particles, etc.) from aqueous emulsions in continuous phase carrier fluids, and especially surfactant-stabilized perfluorocarbon-based oils. In addition, it would be desirable to have the method include a minimum number of steps (in order to minimize any potential damage to cell viability).

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DETAILED DESCRIPTION

The present invention proposes methods for recovering aqueous phase components from aqueous emulsions that have been collected on a microfluidic device in a minimum number of steps and in a gentle manner so as to minimize potential damage to cell viability. Such microfluidic devices are generally known in the art. An example of a preferred microfluidic device is that provided by RainDance Technologies, Inc. ("RDT"), as described in PCT Application No. _____ filed June 1, 2006, entitled
20 "Microfluidic Devices and Methods of Use," PCT Application No. _____ filed June 1, 2006, entitled "Microfluidic Devices and Methods of Use in the Formation and Control of Nanoreactors," and PCT Application No. _____ filed June 1, 2006, entitled "Microfluidic Devices, Methods of Use, and Kits for Performing Diagnostics" each of which are incorporated herein by reference in their entirety.

25 Generally, the methods disclosed herein allow for quantitative recovery of the material enclosed in aqueous phase droplets that have been emulsified in a continuous phase carrier fluid, such as those collected on a microfluidic device.

In one aspect, the invention provides methods in which a stable aqueous emulsion containing aqueous phase components in a continuous phase carrier fluid is allowed to
30 cream to the top of the continuous phase carrier oil. By way of nonlimiting example, the continuous phase carrier fluid can include a perfluorocarbon oil that can have one or more stabilizing surfactants. The aqueous emulsion rises to the top or separates from the continuous phase carrier fluid by virtue of the density of the continuous phase fluid being

greater than that of the aqueous phase emulsion. For example, the perfluorocarbon oil used in one embodiment of the device is 1.8, compared to the density of the aqueous emulsion, which is 1.0.

5 The creamed emulsion is then placed onto a second continuous phase carrier fluid which contains a de-stabilizing surfactant (*e.g.*, 1*H*,1*H*,2*H*,2*H*-Perfluoro-1-octanol). The second continuous phase carrier fluid can also be a perfluorocarbon oil.

Upon mixing, the aqueous emulsion begins to coalesce, and coalescence is completed by brief centrifugation at low speed (*e.g.*, 1 minute at 2000 rpm in a microcentrifuge). The coalesced aqueous phase can now be removed (cells can be placed
10 in an appropriate environment for growth or analysis).

Additional destabilizing surfactants and/or oil combinations can be identified or synthesized to be useful with this invention.

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DESTABILIZING EMULSIONS ON MICROFLUIDIC DEVICES

ABSTRACT

Methods for destabilizing an aqueous phase emulsion in a continuous phase carrier fluid by adding a destabilizing surfactant are described herein.

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