



Method for Isolating Weakly Interacting Molecules from a Fluidic Sample

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The Wisconsin Alumni Research Foundation (WARF) is seeking commercial partners interested in developing a method to isolate weakly interacting molecules from a complex mixture using an immiscible phase filtration technique.

Overview

Molecule-molecule interactions, including interactions between proteins, nucleic acids and other small molecules, are important in a variety of cellular events. A number of physical, molecular biological and genetic methods have been developed to isolate and identify molecular interactions.

Methods such as protein affinity chromatography and immunoprecipitation (IP) require a series of washing steps to remove unbound protein, nucleic acids, cell debris and residual lysis buffer after isolating the proteins, which may impede downstream analyses such as mass spectroscopy. Multistep solid phase extraction may be used to isolate and purify nucleic acids by binding nucleic acids to an immobilized solid phase, then using repeated washes to remove contaminants. High-throughput versions of the solid phase extraction process are available, but are labor intensive and can require expensive robotics, which limits widespread adoption.

Recently, researchers have developed microfluidic embodiments of sample preparation, which can provide practical advantages such as reduced reagent consumption, increased automation, lower device cost and enhanced throughput. However, these methods have not offered significant improvements or reduced complexity over existing methods of isolating proteins and other molecules. Multiple washing steps can inadvertently wash away weakly interacting molecules and add to microfluidic engineering complexity. A need exists for an improved method of sample preparation that addresses the challenge of separating weakly interacting molecules and overcomes the limitations of existing methods.

The Invention

UW-Madison researchers have developed a method for isolating weakly interacting molecules from a fluidic sample using an immiscible phase filtration technique. A mixture is formed using a fluidic sample and a solid phase substrate including at least one immobilized molecule. The mixture is incubated under conditions that allow the immobilized molecule to interact with a target molecule in the fluidic sample to form a solid phase, substrate-immobilized molecule-target molecule complex. The complex is immediately transferred into an immiscible phase by applying an external force to the solid phase substrate.

Immiscible phase filtration allows for the isolation and identification of weakly interacting molecules from the fluidic sample that were previously unidentifiable using traditional methods, as the analyte is isolated very quickly when the solid phase enters the immiscible phase. The method also provides the capability of providing a “snapshot” of the molecular interactions at equilibrium, which is not possible in traditional methods requiring multiple washes.

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Applications

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- Isolation of weakly interacting molecules for experiments including enzyme kinetics, discovery of weakly bound co-regulators and sensing of weakly bound analytes
- Purification of a wide variety of DNA, RNA, proteins and cells from a variety of complex matrices

Key Benefits

- Eliminates need for complex and time-consuming washing steps
- Increases amount of weakly bound molecules captured in sample preparation
- Allows isolation and identification of weakly interacting molecules not possible with traditional methods
- Provides capability of taking a “snapshot” of molecular interactions at equilibrium

Additional Information

For More Information About the Inventors

- [David Beebe](#)

Related Technologies

- [For more information about a microfluidic device for simplified sample purification, see WARF reference number P100050US01.](#)

Tech Fields

- [Analytical Instrumentation, Methods & Materials : Microfluidics](#)
- [Research Tools : DNA & RNA tools](#)

For current licensing status, please contact Jeanine Burmania at jeanine@warf.org or 608-960-9846

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