

What is claimed is:

1. A method for accurate diagnosis of a disease, comprising the steps of:

- (a) providing a sample solution comprising a target biomarker of said disease;
- (b) providing a porous material embedded with components of an aqueous two-phase system (ATPS), wherein said components form two phase solutions when an aqueous solution travels through said porous material so that one phase solution travels faster than the other phase solution;
- (c) allowing said porous material to wick said sample solution, wherein the target biomarker in the sample solution partitions into the faster moving phase solution and becomes concentrated at the front of the faster moving phase solution;
- (d) collecting the target biomarker from the front of the faster moving phase solution, resulting in a final solution containing the target biomarker at a concentration at least 50-fold higher than the concentration of the target biomarker in said sample solution; and
- (e) subjecting said final solution to a diagnostic assay for detection or quantification of said target biomarker;

wherein the accuracy of diagnosing said disease is significantly improved.

2. The method of claim 1, wherein the porous material is selected from the group consisting of fiber-glass paper, cotton-based paper, single-layer matrix paper and polyolefin foam pad.

3. The method of claim 1, wherein said ATPS components are selected from polymers, salts and surfactants.

4. The method of claim 3, wherein said polymers are selected from the group consisting of polyalkylene glycols, poly(oxyalkylene)polymers, poly(oxyalkylene)copolymers, polyvinyl pyrrolidone, polyvinyl alcohol, polyvinyl caprolactam, polyvinyl methylether, alkoxyated surfactants, alkoxyated starches, alkoxyated cellulose, alkyl hydroxyalkyl cellulose, silicone-modified polyethers, poly N-isopropylacrylamide, polyethylene glycol, polypropylene glycol and dextran.

5. The method of claim 3, wherein said salts are selected from the group consisting of kosmotropic salts, chaotropic salts, inorganic salts having a cation of trimethyl ammonium, triethyl ammonium, tripropyl ammonium, tributyl ammonium, tetramethyl ammonium,

tetraethyl ammonium, tetrapropyl ammonium or tetrabutyl ammonium, and an anion of phosphate, sulphate, nitrate, chloride or hydrogen carbonate, NaCl, Na₃PO₄, K₃PO₄, Na₂SO₄, potassium citrate, (NH₄)₂SO₄, sodium citrate, sodium acetate, ammonium acetate, and any combinations thereof.

5

6. The method of claim 3, wherein said surfactants are selected from the group consisting of nonionic surfactants, detergents, Triton-X, Igepal CA-630 and Nonidet P-40.

10

7. The method of claim 1, wherein the sample solution comprises one or more of blood, plasma, serum, urine, saliva, fecal matters and bodily discharges.

8. The method of claim 1, wherein the target biomarker is selected form the group consisting of nucleic acids, proteins, carbohydrates, lipids, metal ions, vitamins and metabolites.

15

9. The method of claim 8, wherein the nucleic acids are cell-free DNA or circulating tumor DNA.

20

10. The method of claim 1, wherein the target biomarker is selected form the group consisting of alpha-fetoprotein (AFP), Cancer Antigen (CA15-3), cancer Antigen (CA125), Prostate specific antigen BRCA1, Carcinoembryonic antigen, c-erb-2, P16, p53, Aldose reductase, Angiogenin, Annexin A1, B-cell activating factor (BAFF), B-cell lymphoma 2 (BCL2)-like 2, Beta Human chorionic gonadotropin, Ca15-3, Calcyclin, Calvasculin, Cathepsin D, Caveolin-1, Chromogranin A, Alpha-crystallin B chain (CRYAB), Endostatin, Eotaxin-2, Epithelial cell adhesion molecule (EpCAM), Ezrin, fatty acid binding protein 4 (FABP4), Galectin-3, γ -glutamylcysteine ligase regulatory chain (GCLR), Gelsolin, Glucose 6-phosphate (G6P), Glycoprotein 130 (gp130), Glutathione S-transferase Mu 1 (GSTM1), Hepsin, High-mobility group protein B1 (HMGB-1), Insulin-like growth factor binding protein 1 (IGFBP-1), Insulin-like growth factor binding protein 4 (IGFBP-4), Insulin-like growth factor binding protein 5 (IGFBP-5), Insulin-like growth factor binding protein 6 (IGFBP-6), LGL, latency associated peptide (LAP), macrophage stimulating protein (MSP), MHC class I polypeptide-related sequence A (MICA), Nucleoside diphosphate kinase B (NME2), Neuron-specific Enolase (NSE), Osteopontin, Osteoprotegerin, Pepsinogen, Peroxiredoxin, Phosphoserine aminotransferase (PSAT1), Prostate Specific Antigen, Receptor tyrosine-protein kinase erbB-3 (ErbB3), Serpin B3, Vascular smooth muscle cell growth factor R2 (VSGF R2/KDR), Vascular endothelial growth factor R3 (VEGF R3/Et-

35

4), Thyroglobulin, Tyrosine kinase with immunoglobulin-like and EGF-like domains 2 (TIE-2), Tissue plasminogen activator (tPA), Transforming growth factor beta (TGF- β 1), Tumor necrosis factor receptor 1 (TNF-R1), urokinase-type Plasminogen Activator (uPA), urokinase-type Plasminogen Activator Receptor (uPAR), BrcaI, BrcaII, kallikreins, e-cadherin, Hox peptide, and Engrailed-2.

11. The method of claim 1, wherein the target biomarker is concentrated by up to 100-fold.

12. The method of claim 1, wherein the disease is cancer.

13. The method of claim 1, wherein the diagnostic assay is selected from the group consisting of Enzyme-Linked Immune Sorbent Assay (ELISA), Polymerase Chain Reaction (PCR), Radioimmunoassay (RIA), proteomic-based approach, blotting procedure, immunoassay, enzymatic assay and GC/MS.

Claims

What is claimed is:

1. A method for accurate diagnosis of a disease, comprising the steps of:
 - (a) providing a sample solution comprising a target biomarker of said disease;
 - (b) providing a porous material embedded with components of an aqueous two-phase system (ATPS), wherein said components form two phase solutions when an aqueous solution travels through said porous material so that one phase solution travels faster than the other phase solution;
 - (c) allowing said porous material to wick said sample solution, wherein the target biomarker in the sample solution partitions into the faster moving phase solution and becomes concentrated at the front of the faster moving phase solution;
 - (d) applying a washing buffer on the ATPS to wash off impurities from said porous material, wherein the target biomarker is retained in the porous material;
 - (e) drying and storing said porous material containing the target biomarker;
 - (f) eluting the target biomarker from the front of the faster moving phase solution by an elution buffer, resulting in a final solution containing the target biomarker at a concentration at least 50-fold higher than the concentration of the target biomarker in said sample solution; and
 - (g) subjecting said final solution to a diagnostic assay for detection and quantification of said target biomarker;

wherein the accuracy of diagnosing said disease is significantly improved; and

wherein the target biomarker is cell-free DNA;

wherein said ATPS components comprise polymers, salts and surfactants; and

wherein said salts are selected from the group consisting of kosmotropic salts, chaotropic salts, inorganic salts having a cation of trimethyl ammonium, triethyl ammonium, tripropyl ammonium, tributyl ammonium, tetramethyl ammonium, tetraethyl ammonium, tetrapropyl ammonium or tetrabutyl ammonium, and an anion of phosphate, sulphate, nitrate, chloride or

hydrogen carbonate, NaCl, Na₃PO₄, K₃PO₄, Na₂SO₄, potassium citrate, (NH₄)₂SO₄, sodium citrate, sodium acetate, ammonium acetate, and any combinations thereof.

2. The method of claim 1, wherein the porous material is selected from the group consisting of fiber-glass paper, cotton-based paper, single-layer matrix paper and polyolefin foam pad.
3. The method of claim 1, wherein said polymers are selected from the group consisting of polyalkylene glycols, poly(oxyalkylene)polymers, poly(oxyalkylene)copolymers, polyvinyl pyrrolidone, polyvinyl alcohol, polyvinyl caprolactam, polyvinyl methylether, alkoxylated surfactants, alkoxylated starches, alkoxylated cellulose, alkyl hydroxyalkyl cellulose, silicone-modified polyethers, poly N-isopropylacrylamide, polyethylene glycol, polypropylene glycol and dextran.
4. The method of claim 1, wherein said surfactants are selected from the group consisting of nonionic surfactants, detergents, Triton-X, Igepal CA-630 and Nonidet P-40.
5. The method of claim 1, wherein the sample solution comprises one or more of blood, plasma, serum, urine, saliva, fecal matters and bodily discharges.
6. The method of claim 1, wherein the target biomarker is concentrated by up to 100-fold.
7. The method of claim 1, wherein the disease is cancer.
8. The method of claim 1, wherein the diagnostic assay is selected from the group consisting of Enzyme-Linked Immune Sorbent Assay (ELISA), Polymerase Chain Reaction (PCR), Radioimmunoassay (RIA), proteomic-based approach, blotting procedure, immunoassay, enzymatic assay and GC/MS.