WHAT IS CLAIMED:

1. A method of fractionating a sample, comprising:
    a) introducing a sample into a porous material;
    b) optionally centrifuging the porous material containing the sample;
    c) drying the sample in the porous material;
    d) extracting a first set molecules from the dried sample-containing porous material, comprising:
        1) incubating the dried sample-containing porous material in a first extraction solution;
        2) separating the first extraction solution from the porous material; and
        3) optionally, washing the porous material;
    e) digesting proteins remaining in the porous material, comprising:
        1) incubating the porous material in a digestion solution; and
        2) separating the digestion solution from the porous material;
    f) detecting one or more molecules in the separated first extraction solution; and
    g) detecting one or more peptides or proteins in the digestion solution.

2. The method of claim 1, wherein following extraction of the first set of molecules and prior to the digestion step the method further comprises:
    A) extracting a second set of molecules from the first extracted porous material, comprising:
        1) incubating the first extracted porous material in a second extraction solution;
        2) separating the second extraction solution from the second incubated porous material; and
        h) optionally, washing the separated second extracted porous material; and
    B) optionally, detecting the second set of molecules in the separated second extraction solution.

3. The method of claim 2, wherein prior to the digestion step the method further comprises sequentially extracting one or more further sets of molecules from the second extracted porous material comprising repeating the steps A)-B) with one or more further extraction solutions.

4. The method of claim 3, wherein the first set of molecules, the second set of molecules, or the one or more further sets of molecules is selected from the group consisting of proteins, phosphoproteins, phosphopeptides, nucleic acids, and glycans, preferably wherein said proteins in the first, second, or one or more further sets of molecules, respectively, and/or the one or more peptides or proteins in the digestion solution, are phosphoproteins or phosphopeptides, more preferably wherein the method comprises

performing an enrichment step to enrich for phosphorylated proteins and/or peptides in the separated first, second, or one or more extraction solutions and/or the digestion solution.

5. The method of claim 1-4, wherein the extraction solution comprises an extracting agent comprising a salt, a mild detergent, a strong detergent, a chaotrope, a reducing agent, a thiol-containing reducing agent, or an alkylating agent, an acid, an organic solvent, or an enzyme, or combinations thereof; and/or

wherein the digestion solution comprises a reducing agent, an alkylating agent, a buffer, a detergent, or combinations thereof, and optionally, wherein the digestion solution further comprises a salt, a mild detergent, a strong detergent, a chaotrope, or a thiol-containing reducing agent, an acid, an organic solvent, or an enzyme, or combinations thereof;

preferably, wherein:

- the salt is or comprises NaCl, LiCl, $CaCl_2$, or Tris-HCl;
- the mild detergent is a non-ionic detergent, an ionic detergent, or a zwitterionic detergent, optionally wherein the mild detergent is or comprises: i) a non-ionic detergent comprising PBS or Tween; ii) an ionic detergent comprising sodium deoxycholate; or iii) a zwitterionic detergent comprising a sulfobetaine or an amidosulfobetaine;
- the strong detergent is or comprises CTAB, CHAPS, or SDS.
- the chaotrope is or comprises urea, thiourea, or guanidine;
- the reducing agent is or comprises a phosphine, optionally wherein the phosphine is or comprises TBP or TCEP;
- the thiol-containing reducing agent is or comprises betamercaptoethanol or dithiothreitol;
- the alkylating agent is or comprises chloroacetamide, iodoacetamide or acrylamide;
- the acid is or comprises citric acid or trifluoroacetic acid;
- the organic solvent is or comprises methanol; and/or
- the organic enzyme is or comprises benzonase;

more preferably, wherein:

- the concentration of the salt in the extraction solution, is 0.01-1 M, more preferably wherein the salt comprises LiCl and Tris-HCl, even more preferably wherein the concentration of LiCl is 0.5 M, and the concentration of Tris-HCl is 0.1 M;
- the concentration of the chaotrope in the extraction solution is 0.01-8 M; and/or
- the extraction solution, comprises urea at a concentration of 2, 5, 6, or 7M, and thiourea;

optionally, wherein the extraction solution comprises an endoglycosidase, preferably wherein the endoglycosidase is Peptide-*N*-Glycosidase F (PNGase F) and/or *O*-glycosidase;

and preferably, wherein the digestion solution comprises:

- triethylammonium bicarbonate, sodium deoxycholate (SDC), TCEP, or chloroacetamide, or combinations thereof; optionally wherein the triethylammonium bicarbonate is present in the digestion solution at a concentration of 0.1 M, wherein the sodium deoxycholate (SDC) is present in the digestion solution at a concentration of 0.05% to 10% (w/v), wherein the TCEP is present in the digestion solution at a concentration of 1 mM to 100 mM, and/or wherein the chloroacetamide is present in the digestion solution at a concentration of 5 mM to 100 mM; and/or
- a protease or a combination of proteases, preferably, wherein the protease is trypsin or the combination of proteases comprises trypsin.

6. The method of claim 4 or 5, wherein:
- the first, the second, the one or more further extraction solutions, and the digestion solutions are different;
- selection of the first, the second, the one or more further extraction solutions is to fractionate the proteins contained in the porous material via differential solubility;
- each sequential extraction solution selected is characterized as having greater solubility of the proteins remaining in the porous material; or
- the first, the second, the third, and the further extraction solutions comprise identical extracting agents of increasing concentrations as progress from one extraction to the next extraction.

7. The method of claim 1-6, wherein the sample is or comprises: a body fluid sample, cells or tissue, cells suspended in a liquid, cultured cells suspended in a culture media, blood, optionally wherein the blood sample is: a whole blood (WB) sample, a red blood cell (RBC) sample, a white blood cell (WBC) sample, a frozen blood sample, or a fresh blood sample, blood fractions, plasma, urine, tears, wound fluid, CSF, bronchoalveolar lavage, or ascites, preferably, wherein the sample has a volume in the range of 100 µL to 2 µL, 100 µL to 5 µL, 100 µL to 10 µL, 99 µL to 2 µL, 90 µL to 2 µL, 80 µL to 2 µL, 70 µL to 2 µL, 60 µL to 2 µL, 50 µL to 2 µL, 40 µL to 2 µL, 30 µL to 2 µL, 20 µL to 2 µL, 10 µL to 2 µL, or 5 µL to 2 µL.

8. The method of any one of claim 1-7, wherein the introducing step comprises absorbing the sample into the porous material, preferably by absorbing the sample into the porous material via a finger prick, dipping the porous material into the sample, or pipetting a known volume of the sample into the porous material.

9. The method of any one of claim 1-8, comprising drying the sample in the porous material and wherein the drying step comprises drying for a period of time to adhere the sample to the porous material, optionally wherein the drying step comprises:

- air drying the sample within the porous material, wherein the drying step is for less than 30 minutes, preferably at least 5 minutes, or wherein the drying is air drying is for at least 1, 2, 4, 6, 8, 10, 12, 16, 20 or 24 hours, optionally wherein the drying is in the presence of a desiccant,
- centrifuging the sample within the porous material, preferably wherein the centrifuging is at a speed of 500 g to 10,000 g, more preferably for at least 1 to 15 minutes or for no more than 15 minutes; and/or
- vacuuming the sample within the porous material.

10. The method of any one of claim 1-9, further comprising storing the sample-containing porous material for a period of time prior to the first extraction step, optionally, wherein
storing the sample-containing porous material immediately follows the drying step;
the sample-containing porous material is stored frozen; or
the sample-containing porous material is stored at room temperature,
preferably, wherein the drying step, the extraction step, and the digesting step are conducted with the sample-containing porous material in a tube, more preferably, wherein the extraction solution has a volume of less than 100 µL, a volume less than 95 µL, a volume less than 90 µL, a volume of 1-5 times the volume of porous material, a volume of 1, 2, 3, 4, or 5 times the volume of porous material, even more preferably a volume of 3 times the volume of porous material.

11. The method of any one of claim 1-10, wherein:

a) the incubating in the extraction solution is conducted for a period of time long enough to extract one or more proteins contained within or adhered to the porous material; optionally wherein said incubating is conducted for 1, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, or 60 minutes, or wherein said incubating is conducted for 1-48 hours, 1-42 hours, 1-36 hours, 1-30 hours, 1-24 hours, 1-18 hours, 1-12 hours, or 1-6 hours; wherein said incubating is conducted at ambient temperature, or wherein said incubating is conducted at a temperature elevated above ambient temperature.

b) the incubating in the digestion solution is conducted for a period of time long enough to digest one or more proteins contained within or adhered to the porous material, optionally wherein said incubating is conducted for 1 minute - 48 hours, wherein said incubating is conducted for 1, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55 or 60 minutes, or wherein said incubating is conducted for 1, 2, 3,

4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23 or 24 hours, and/or said incubating is conducted at ambient temperature, or conducted at a temperature elevated above ambient temperature, preferably at 37°C; and /or wherein said incubating is conducted with agitation; and/or

c) wherein the method comprises washing the separated extracted porous material, optionally wherein the separated extracted porous material is washed with a washing volume of the extraction solution, a volume of an extraction solution different from the extraction solution, or in a volume of water, wherein selection of the different extraction solution is limited to solutions based on proteins remaining in the separated extracted porous material that have equivalent or less solubility in the different extraction solution than the preceding extraction solution, and wherein the washing volume of the extraction solution is between 5 times the volume of porous material and 3 mL, wherein the washing volume of the extraction solution is 0.05-3 mL, and/or wherein the washing step is repeated 1, 2, or 3 times.

12. The method of any one of claim 1-11, wherein said detecting one or more molecules in the separated first extraction solution comprises:

A) detecting proteins in the separated first extraction solution, optionally wherein

- the detecting of proteins is via immunoassay, a Western blot, or ELISA;
- the detecting step comprises a Luminex and Proximity Extension Assay, optionally wherein the Luminex and Proximity Extension Assay comprises Slow Offrate Modified Aptamer (SOMAmer) reagents, and/or
- the detecting of proteins is via Mass Spectrometry (MS), optionally wherein the detection via MS comprises subjecting the extraction solution to digestion prior to said detection via Mass Spectrometry (MS); or

B) detecting one or more molecules in the separated first extraction solution comprises detecting glycans in the separated first extraction solution, optionally wherein the detecting of glycans is via High Performance Liquid Chromatography (HPLC);

and/or wherein the detecting of one or more peptides or proteins in the digestion solution is via Mass Spectrometry (MS);

preferably, wherein said MS is LC-MS, selected reaction monitoring mass spectrometry (SRM-MS), data-dependent acquisition MS (DDA-MS), data-independent acquisition MS (DIA-MS), or is selected from the group consisting of: matrix-assisted laser desorption/ionisation time-of-flight (MALDI-TOF) MS; MALDI-TOF post-source-decay (PSD); MALDI- TOF/TOF; surface-enhanced laser desorption/ionization

time-of-flight mass spectrometry (SELDI-TOF) MS; electrospray ionization mass spectrometry (ESI-MS); ESI-MS/MS; ESI-MS/(MS)n (n is an integer greater than zero); ESI 3D or linear (2D) ion trap MS; ESI triple quadrupole MS; ESI quadrupole orthogonal TOF (Q-TOF); ESI Fourier transform MS systems; desorption/ionization on silicon (DIOS); secondary ion mass spectrometry (SIMS); atmospheric pressure chemical ionization mass spectrometry (APCI-MS); APCI- MS; APCI- (MS)n; ion mobility spectrometry (IMS); inductively coupled plasma mass spectrometry (ICP-MS); atmospheric pressure photoionization mass spectrometry (APPI- MS), APPI-MS/MS; and APPI- (MS)n.

13. The method of any one of claim 1-12, wherein the porous material is a hydrophilic polymeric material, optionally wherein the porous material comprises a plastic, and/or the porous material comprises a sponge, or is an absorbent tip of a volumetric absorptive microsampling (VAMS) device, optionally wherein the method uses a plurality of the porous materials, wherein, optionally, the porous material is pre-loaded with

i) a protease inhibitor;

ii) an enzyme, preferably a nuclease, more preferably benzonase;

iii) a detergent, preferably SDS; or

iv) an anticoagulant; preferably wherein the anticoagulant is EDTA or heparin.

14. The method of any one of claim 1-13, wherein

one or more proteins detected in a set of molecules or in the separated digestion solution is or comprises a non-membrane/soluble protein complex; and/or

one or more proteins detected in set of molecules or in the separated digestion solution is or comprises a cell-membrane associated protein, optionally wherein the cell-membrane associated protein is a cell-membrane bound protein, a cell integral-membrane protein or a membrane protein complex; and/or

the method recovers from a red blood cell (RBC) sample, a plasma sample, or a whole blood sample, a fraction of non-membrane/soluble protein complexes not obtained by traditional microsampling, or a fraction of membrane protein complexes not obtained by traditional microsampling analysis.

15. The method of claim 1, wherein

i) the porous material is a hydrophilic polymer tip of a volumetric absorptive microsampling (VAMS) device, wherein the first extraction solution comprises 100mM-500mM LiCl and 100mM Tris, and wherein the method comprises washing the porous material at step d) 3) with a wash solution comprising 100mM-500mM LiCl and 100mM Tris, and wherein the digestion solution comprises

1 µg Trypsin in a buffer comprising 1% sodium deoxycholate (SDC), 10 mM tris (2-carboxyethyl) phosphine (TCEP), 40 mM 2-Chloroacetamide (CLA), 100mM and 100mM triethylammonium bicarbonate buffer (TEAB);

ii) the porous material is a hydrophilic polymer tip of a volumetric absorptive microsampling (VAMS) device, wherein the first extraction solution comprises a PBS solution comprising a protease inhibitor, wherein the volume of the first extraction solution is ≤ 100µL; wherein the method comprises washing the porous material at step d) 3) with a wash solution comprising 100mM-500mM LiCl and 100mM Tris; wherein the digestion solution comprises 1 µg Trypsin in a buffer comprising 1% sodium deoxycholate (SDC), 10 mM tris (2-carboxyethyl) phosphine (TCEP), 40 mM 2-Chloroacetamide (CLA), 100mM and 100mM triethylammonium bicarbonate buffer (TEAB); and wherein the one or more molecules in the separated first extraction solution is one or more proteins and detection is via immunoassay; or

iii) the porous material is a hydrophilic polymer tip of a volumetric absorptive microsampling (VAMS) device, wherein the first extraction solution comprises a PBS solution comprising a protease inhibitor, wherein the volume of the first extraction solution is ≤ 100µL; wherein the method comprises washing the porous material at step d) 3) with a wash solution comprising 100mM-500mM LiCl and 100mM Tris; wherein the digestion solution comprises 1 µg Trypsin in a buffer comprising 1% sodium deoxycholate (SDC), 10 mM tris (2-carboxyethyl) phosphine (TCEP), 40 mM 2-Chloroacetamide (CLA), 100mM and 100mM triethylammonium bicarbonate buffer (TEAB); and wherein the one or more molecules in the separated first extraction solution is one or more proteins and detection is via immunoassay, and wherein following extraction of the first set of molecules and prior to the digestion step the method further comprises:

A) extracting a second set of molecules from the first extracted porous material, comprising:

1) incubating the first extracted porous material in a second extraction solution;

2) separating the second extraction solution from the second incubated porous material; and

h) optionally, washing the separated second extracted porous material; and

B) optionally, detecting the second set of molecules in the separated second extraction solution; wherein the second extraction solution comprises PNGase F in a buffer comprising 1% sodium deoxycholate (SDC), 10 mM tris (2-carboxyethyl) phosphine (TCEP), 40 mM 2-Chloroacetamide (CLA), 100mM and 100mM triethylammonium bicarbonate buffer (TEAB); and wherein the method comprises detecting the second set of molecules in the separated second extraction solution, wherein the one or more molecules is one or more glycans.

16. The method of claim 3, wherein the one or more further extraction solution comprises PNGase F in a buffer comprising 1% sodium deoxycholate (SDC), 10 mM tris (2-carboxyethyl) phosphine (TCEP), 40 mM 2-Chloroacetamide (CLA), 100mM and 100mM triethylammonium bicarbonate buffer (TEAB); and wherein the method comprises detecting a further set of molecules in the separated further extraction solution, wherein the one or more molecules is one or more glycans, preferably wherein the method, further comprises dividing the porous material prior to the digestion step e), and analysing a portion of the portion of the porous material for remaining glycans.

17. A method of producing a protein profile, comprising the method of any one of claim 1-16, optionally wherein the protein profile is produced by immunoassay work flow, a proteomics work flow, or a combination of an immunoassay work flow and a proteomics work flow; and wherein the method produces a protein profile of non-membrane/soluble protein complexes, and/or the method produces a protein profile of cell-membrane associated proteins.

18. A method of producing a protein profile comprising:

a) obtaining at least one protein profile produced according to one or more of claim 1-17 from a sample that has been obtained from a subject having a disease or disorder;
b) obtaining at least one protein profile produced according to one or more of claims 1-17 from a sample that has been obtained from at least one subject not having the disease or disorder;
c) comparing the protein profile of the subject having the disease or disorder to the protein profile of the at least one subject not having the disease or disorder, and
d) producing the disease protein profile from the comparison, wherein said produced disease protein profile comprises one or more proteins that have a different presence or level in the protein profile from the subject having the disease or disorder compared to the protein profile of the at least one subject not having the disease or disorder.