

What is claimed is:

1. A method for characterizing host-cell proteins in a sample matrix, comprising:
enriching host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support;
performing fractionation on a flowthrough from the affinity chromatography; and
characterizing at least one of the host-cell proteins using a mass spectrometer.
2. The method of claim 1, wherein the affinity chromatography support is protein A chromatography support.
3. The method of claim 1 further comprising washing the affinity chromatography support with a wash buffer and collecting the flow-through.
4. The method of claim 1, wherein the affinity chromatography support comprises protein A or protein G.
5. The method of claim 4, wherein the protein A or the protein G is immobilized on agarose or sepharose resin.
6. The method of claim 1, wherein the mass spectrometer is a tandem mass spectrometer.
7. The method of claim 6, wherein the mass spectrometer is coupled with a liquid chromatography system.
8. The method of claim 7, wherein the liquid chromatography system is a nano-liquid chromatography system.
9. The method of claim 1 further comprising characterizing at least one of the host-cell proteins using High-Field Asymmetric Waveform Ion Mobility Spectrometry device.
10. The method of claim 1, wherein the sample matrix further comprises a protein of interest.
11. The method of claim 10, wherein the protein of interest is an antibody.
12. The method of claim 10, wherein the protein of interest is a fusion protein.
13. The method of claim 1, wherein the fractionation is a size-based fractionation.
14. The method of claim 1, wherein the fractionation is a hydrophobicity-based fractionation.

15. The method of claim 1, wherein fractionation is a charge-based fractionation.
16. The method of claim 1, wherein fractionation is a pI-based fractionation.
17. The method of claim 1, wherein the fractionation comprises fractionation by liquid chromatography.
18. The method of claim 17, wherein the liquid chromatography is reversed phase liquid chromatography.
19. The method of claim 1, wherein the method is capable of characterizing at least about 50% more host-cell proteins than a method that enriches host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support without performing the fractionation step.
20. The method of claim 1, wherein the method is capable of characterizing at least about 50% more host-cell proteins than a method that performs a fractionation without enriching host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support.
21. The method of claim 1, wherein the method is capable of characterizing at least about 50% to about 75% more host-cell proteins than a method that enriches host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support without performing the fractionation step.
22. The method of claim 1, wherein the method is capable of characterizing at least about 50% to about 75% more host-cell proteins than a method that performs a fractionation without enriching host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support.
23. A method for characterizing host-cell proteins in a sample matrix having a protein of interest, comprising:
enriching host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support;
washing the affinity chromatography support with a wash buffer;
collecting a flow-through;

- performing fractionation on a sample obtained after performing the enrichment step; and characterizing at least one of the host-cell proteins using a mass spectrometer.
24. The method of claim 23, wherein the flow-through has a reduced amount of protein of interest than the sample matrix.
25. A method for characterizing host-cell proteins in a sample matrix, comprising:
enriching host-cell proteins in said mixture by contacting the sample matrix with an affinity chromatography support to obtain a mixture;
subjecting the mixture to non-denaturing digestion conditions; and
characterizing at least one of the host-cell proteins using a mass spectrometer.
26. The method of claim 25, wherein the affinity chromatography support is protein A chromatography support.
27. The method of claim 25 further comprising collecting the flow-through from the affinity chromatography support.
28. The method of claim 25, wherein the affinity chromatography support comprises protein A or protein G.
29. The method of claim 28, wherein the protein A or the protein G is immobilized on agarose or sepharose resin.
30. The method of claim 25, wherein the mass spectrometer is a tandem mass spectrometer.
31. The method of claim 25, wherein the mass spectrometer is coupled with a liquid chromatography system.
32. The method of claim 31, wherein the liquid chromatography system is a nano-liquid chromatography system.
33. The method of claim 25, wherein the mass spectrometer is a High-Field Asymmetric Waveform Ion Mobility Spectrometer.
34. The method of claim 28, wherein the sample matrix further comprises a protein of interest.
35. The method of claim 34, wherein the protein of interest is at least one selected from the

group consisting of an antibody or a fragment or derivative thereof, a fusion protein, and a physiologically active non-antibody protein.

36. The method of claim 35, wherein the method is capable of characterizing at least about 500% more host-cell proteins than a method that subjects the mixture to non-denaturing digestion conditions to form a mixture without enriching host-cell proteins in said mixture by contacting the sample matrix with an affinity chromatography support to obtain a mixture.
37. The method of claim 34, wherein the method is capable of characterizing at least about 100% to about 1000% more host-cell proteins than a method subjects the mixture to non-denaturing digestion conditions to form a mixture without enriching host-cell proteins in said mixture by contacting the sample matrix with an affinity chromatography support to obtain a mixture.
38. A method for characterizing host-cell proteins in a sample matrix, comprising:
enriching host-cell proteins in the sample matrix by contacting the sample matrix with an affinity chromatography support; and
characterizing at least one of the host-cell proteins using a High-Field Asymmetric Waveform Ion Mobility Spectrometry.
39. The method of claim 38, wherein the method is capable of characterizing at least about 30% more host-cell proteins than a method not comprising a High-Field Asymmetric Waveform Ion Mobility Spectrometry.
40. The method of claim 38, wherein the method is capable of characterizing at least about 30% to about 75% more host-cell proteins than a than a method not comprising a High-Field Asymmetric Waveform Ion Mobility Spectrometry.
41. A method for characterizing host-cell proteins in a sample matrix, comprising:
enriching the host-cell proteins in the sample matrix by contacting a sample matrix with an affinity chromatography support to obtain a mixture;
subjecting the mixture to non-denaturing digestion conditions; and
characterizing of at least one of the host-cell proteins using a High-Field Asymmetric

Waveform Ion Mobility Spectrometry.

42. The method of claim 41, wherein the method is capable of characterizing at least about 15% more host-cell proteins than a method that enriches the host-cell proteins in the sample matrix by contacting a sample matrix with an affinity chromatography support to obtain a mixture and subjects the mixture to non-denaturing digestion conditions and characterizing of at least one of the host-cell proteins using a mass spectrometry device other than a High-Field Asymmetric Waveform Ion Mobility Spectrometry device.
43. The method of claim 41, wherein the method is capable of characterizing at least about 15% to about 60% more host-cell proteins than a method that enriches the host-cell proteins in the sample matrix by contacting a sample matrix with an affinity chromatography support to obtain a mixture and subjects the mixture to non-denaturing digestion conditions and characterizing of at least one of the host-cell proteins using a mass spectrometry device other than a High-Field Asymmetric Waveform Ion Mobility Spectrometry device.