What is Claimed is:

1. A method for isolating an antigen from a vaccine composition, the method comprising the steps of:

(a) solubilizing the antigen in the vaccine composition by reduction without a detergent and without an alkylating agent, wherein the reduction comprises an incubation temperature of about 70°C, about 75°C, about 80°C, about 85°C, or about 90°C; and

(b) isolating the antigen or an antigen subtype by fractionation.

2. The method of claim 1, wherein the reduction comprises treating the vaccine composition with dithiothreitol.

3 The method of claim 1 or 2, wherein the solubilizing step further comprises denaturing with a chaotropic agent.

4. The method of any one of claims 1-3, wherein the reduction is further carried out by acidification of the antigen or antigen subtype to prevent disulfide bond formation between separated antigen subtypes.

5. A method for quantifying antigen or antigen subtype in a vaccine composition, the method comprising the method of any one of claims 1-4 with a further step of quantifying the antigen or antigen subtype.

6. The method of claim 5, wherein the quantifying step is carried out without using an antigen standard.

7. The method of claim 5, wherein the quantifying step is carried out using an antigen standard.

8. The method of any one of claims 6-7, wherein the quantifying step comprises quantifying antigen by amino acid analysis.

9. The method of any one of claims 1-8, wherein the reduction comprises an incubation time from about 5 minutes to about 20 hours.

10. The method of claim 9, wherein the reduction comprises an incubation time from about 30 minutes to about 2 hours.

11. The method of claim 10, wherein the reduction comprises an incubation time of about 1 hour.

12. The method of any one of claims 1-11, wherein the reduction comprises an incubation temperature at about 85°C.

13. The method of any one of claims 3-12, wherein the chaotropic agent is guanidine hydrochloride, urea, thiourea, lithium, perchlorate, or thiocyanate.

14. The method of any one of claims 4-13, wherein the acidification is carried out with phosphoric acid, hydrochloric acid, sulfuric acid, trifluoroacetic acid, pentafluoropropionic acid, heptafluorobutyric acid, or formic acid.

15. The method of any one of claims 1-14, wherein fractionation is carried out by chromatography.

16. The method of claim 15, wherein the chromatography is high performance liquid chromatography (HPLC), reversed-phase HPLC (RP-HPLC), ion exchange-HPLC (IEX-HPLC), affinity chromatography, hydrophobic interaction chromatography (HIC), or size exclusion chromatography (SEC).

17. The method of claim 16, wherein the chromatography is RP-HPLC.

18. The method of any one of claims 1-17, wherein the antigen is viral or bacterial.

19. The method of any one of claims 1-18, wherein the antigen is hemagglutinin (HA).

20. The method of any one of claims 1-19, wherein the HA is from an Influenza virus vaccine composition, a measles virus vaccine composition, a parainfluenza virus vaccine composition, or a mumps virus vaccine composition.

21. The method of any one of claims 1-20, wherein the vaccine composition is an Influenza virus vaccine composition.

22. The method of any one of claims 1-21, wherein the Influenza virus vaccine composition provides protection from an Influenza virus selected from the group consisting of Influenza A and Influenza B.

23. The method of any one of claims 1-22, wherein the antigen subtype is any one of Influenza A HA1, HA2, HA3, HA4, HA5, HA6, HA7, HA8, HA9, HA10, HA11, HA12, HA13, HA14, HA15, and HA16 and Influenza B HA.

24. The method of any one of claims 1-23, wherein the antigen subtype is Influenza A HA subtype HA1, HA2, HA3, HA5, HA7, HA9, HA10, or influenza B HA.

25. The method of any one of claims 1-24, wherein the Influenza A HA subtype is HA1.

26. The method of any one of claims 23-25, wherein the Influenza vaccine composition provides protection from an Influenza type selected from the group consisting of Influenza A H1N1, H1N2, H2N2, H3N2, H5N1, H7N1, H7N2, H7N3, H7N7, H9N2, H10N7, and Influenza B.

27. The method of any one of claims 4-26, wherein the method quantifies antigen content with a relative standard deviation (RSD) of less than about 2.5%.

28. The method of claim 27, wherein the RSD is about 2.2%.

29. The method of any one of claims 27-28, wherein the RSD is about 1.3%.

What is Claimed is:

1. A method for isolating an antigen from a vaccine composition, the method comprising the steps of:

(a) solubilizing the antigen in the vaccine composition without a detergent and without an alkylating agent; and

(b) isolating the antigen or an antigen subtype by fractionation.

2. The method of claim 1, wherein the solubilizing step is carried out by reduction.

3. The method of claim 2, wherein the reduction comprises treating the vaccine composition with dithiothreitol.

4. The method of claim 2, wherein the solubilizing step further comprises denaturing with a chaotropic agent.

5. The method of any one of claims 2-4, wherein the reduction is further carried out by acidification of the antigen or antigen subtype to prevent disulfide bond formation between separated antigen subtypes.

6. A method for quantifying antigen or antigen subtype content in a vaccine composition comprising antigen, the method comprising the method of any one of claims 1-5 with a further step of quantifying the antigen or antigen subtype.

7. The method of claim 6, wherein the quantifying step is carried out without using an antigen standard.

8. The method of claim 6, wherein the quantifying step is carried out using an antigen standard.

9. The method of any one of claims 7-8, wherein the quantifying step comprises quantifying antigen by amino acid analysis.

10. The method of any one of claims 3-9, wherein the reduction comprises an incubation time from about 5 minutes to about 20 hours.

11. The method of any one of claims 3-10, wherein the reduction comprises an incubation time from about 30 minutes to about 2 hours.

12. The method of any one of claims 3-11, wherein the reduction comprises an incubation time of about 1 hour.

13. The method of any one of claims 3-12, wherein the reduction comprises an incubation temperature from about 20ºC to about 100ºC.

14. The method of any one of claims 3-13, wherein the reduction comprises an incubation temperature from about 50ºC to about 90ºC.

15. The method of any one of claims 3-14, wherein the reduction comprises an incubation temperature at about 85ºC.

16. The method of any one of claims 4-15, wherein the chaotropic agent is guanidine hydrochloride, urea, thiourea, lithium, perchlorate, or thiocyanate.

17. The method of any one of claims 4-16, wherein the acidification is carried out with phosphoric acid, hydrochloric acid, sulfuric acid, trifluoroacetic acid, pentafluoropropionic acid, heptafluorobutyric acid, or formic acid.

18. The method of any one of claims 1-17, wherein fractionation is carried out by chromatography.

19. The method of claim 18, wherein the chromatography is high performance liquid chromatography (HPLC), reversed-phase HPLC (RP-HPLC), ion exchange-HPLC (IEX-HPLC), affinity chromatography, hydrophobic interaction chromatography (HIC), or size exclusion chromatography (SEC).

20. The method of claim 19, wherein the chromatography is reversed-phase (RP)-HPLC.

21. The method of any one of claims 1-20, wherein the antigen is viral or bacterial.

22. The method of any one of claims 1-21, wherein the antigen is hemagglutinin (HA).

23. The method of claim 1-22, wherein the HA is from an Influenza virus vaccine composition, a measles virus vaccine composition, a parainfluenza virus vaccine composition, or a mumps virus vaccine composition.

24. The method of any one of claims 1-23, wherein the vaccine composition is an Influenza virus vaccine composition.

25. The method of any one of claims 23-24, wherein the Influenza virus vaccine composition provides protection from an Influenza virus selected from the group consisting of Influenza A and Influenza B.

26. The method of any one of claims 1-25, wherein the antigen subtype is any one of Influenza A HA1, HA2, HA3, HA4, HA5, HA6, HA7, HA8, HA9, HA10, HA11, HA12, HA13, HA14, HA15, and HA16 and Influenza B HA.

27. The method of claim 26, wherein the antigen subtype is Influenza A HA subtype HA1, HA2, HA3, HA5, HA7, HA9, HA10, or influenza B HA.

28. The method of claim 27, wherein the Influenza A HA subtype is HA1.

29. The method of any one of claims 26-28, wherein the Influenza vaccine composition provides protection from an Influenza type selected from the group consisting of Influenza A H1N1, H1N2, H2N2, H3N2, H5N1, H7N1, H7N2, H7N3, H7N7, H9N2, H10N7, and Influenza B.

30. The method of any one of claims 5-29, wherein the method quantifies antigen content with a relative standard deviation (RSD) of less than about 2.5%.

31. The method of claim 29, wherein the RSD is about 2.2%.

32. The method of any one of claims 30-31, wherein the RSD is about 1.3%.