modifying agent by spraying. In another embodiment, the improvement is realized by adjusting the pH of the solution after addition of the alcohol. In a related embodiment, the improvement is realized by adjusting the pH of the solution during addition of the alcohol. In another embodiment, the improvement is realized by increasing the concentration of alcohol (*e.g.*, ethanol) to about 25% (v/v). In another embodiment, the improvement is realized by lowering the temperature of the precipitation step to between about -7°C and -9°C. In a preferred embodiment, the improvement is realized by increasing the concentration of alcohol (*e.g.*, ethanol) to about 25% (v/v) and lowing the temperature to between about -7°C and -9°C. In comparison, both Cohn *et al.* and Oncley *et al.* perform precipitation at -5°C and Oncley *et al.* use 20% alcohol, in order to reduce the level of contaminants in the precipitate. Advantageously, the methods provided herein allow for maximal IgG yield without high levels of contamination in the final product.

[0250] It has been discovered that when the pH of the solution is adjusted to a pH of about 6.9 prior to addition of the precipitating alcohol, the pH of the solution shift from 6.9 to between about 7.4 and about 7.7, due in part to protein precipitation ~~(*see*, Figure 8)~~. As the pH of the solution shifts away from 6.9, precipitation of IgG becomes less favorable and the precipitation of certain contaminants becomes more favorable. Advantageously, the inventors have found that by adjusting the pH of the solution after addition of the precipitating alcohol, that a higher percentage of IgG is recovered in the Fraction II+III precipitate.

[0251] Accordingly, in one aspect, the improvement relates to a method in which a reduced amount of IgG is lost in the supernatant fraction of the modified Fraction II+III precipitation step. In other words, an increased percentage of the starting IgG is present in the Fraction II+III precipitate. In certain embodiments, the process improvement is realized by adjusting the pH of the solution to between about 6.7 and about 7.1 immediately after or during the addition of the precipitating alcohol. In another embodiment, the process improvement is realized by maintaining the pH of the solution to between about 6.7 and about 7.1 continuously during the precipitation incubation period. In other embodiments, the pH of the solution is adjusted to between about 6.8 and about 7.0 immediately after or during the addition of the precipitating alcohol, or to a pH of about 6.7, 6.8, 6.9, 7.0, or 7.1 immediately after or during the addition of the precipitating alcohol. In a particular embodiment, the pH of the solution is adjusted to about 6.9 immediately after or during the addition of the precipitating alcohol. In certain embodiments, the pH of the solution is maintained at between about 6.8 to about 7.0

requirements can be met using a buffering agent selected from, for example, acetate, citrate, monobasic phosphate, dibasic phosphate, mixtures thereof, and the like. Suitable buffer concentrations typically range from about 5 to about 100 mM, or from about 10 to about 50 mM, or about 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, or 100 mM buffering agent.

[0258] The extraction buffer will preferably have a conductivity of from about 0.5 $mS \cdot cm^{-1}$ to about 2.0 $mS \cdot cm^{-1}$. For example, in certain embodiments, the conductivity of the extraction buffer will be about 0.5 $mS \cdot cm^{-1}$, or about 0.6, 0.7, 0.8, 0.9, 1.0, 1.1, 1.2, 1.3, 1.4, 1.5, 1.6, 1.7, 1.8, 1.9, or about 2.0 $mS \cdot cm^{-1}$. One of ordinary skill in the art will know how to generate extraction buffers having an appropriate conductivity.

[0259] In one particular embodiment, an exemplary extraction buffer may contain at or about 5 mM monobasic sodium phosphate and at or about 5 mM acetate at a pH of at or about 4.5 ± 0.2 and conductivity of at or about 0.7 to 0.9 mS/cm.

[0260] Generally, the extraction is performed at between about 0°C and about 10°C, or between about 2°C and about 8°C. In certain embodiments, the extraction may be performed at about 0°C, 1°C, 2°C, 3°C, 4°C, 5°C, 6°C, 7°C, 8°C, 9°C, or 10°C. In a particular embodiment, the extraction is performed at between about 2°C and about 10°C. Typically, the extraction process will proceed for between about 60 and about 300 minutes, or for between about 120 and 240 min, or for between about 150 and 210 minutes, while the suspension is continuously stirred. In certain embodiments, the extraction process will proceed for about 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, or about 300 minutes. In a preferred embodiment, the extraction process will proceed for at least 160 minutes with continuous stirring.

[0261] It has been found that employing an extraction buffer containing 5 mM monobasic sodium phosphate, 5 mM acetate, and 0.051% to 0.06% glacial acetic acid (v/v), a substantial increase in the yield increase in the final IgG composition can be obtained without jeopardizing the purity of the final product. ~~The correlation of amount of acetic acid and extraction buffer pH is demonstrated in Figure 9.~~ In a preferred embodiment, the Fraction II+III precipitate is extracted with a paste to buffer ration of at or about 1:15 at a pH of at or about 4.5 ± 0.2.

realized by mixing finely divided silicon dioxide (SiO_2) with the Fraction II+III suspension for at least about 30 minutes.

5. Pretreatment and Filtration of the Modified Fraction II+III Suspension

[0265] In order to remove the non-solubilized fraction of the Modified Fraction II+III precipitate (*i.e.*, the Modified Fraction II+III filter cake), the suspension is filtered, typically using depth filtration. Depth filters that may be employed in the methods provided herein include, metallic, glass, ceramic, organic (such as diatomaceous earth) depth filters, and the like. Example of suitable filters include, without limitation, Cuno 50SA, Cuno 90SA, and Cuno VR06 filters (Cuno). Alternatively, the separation step can be performed by centrifugation rather than filtration.

[0266] Although the manufacturing process improvements described above minimize IgG losses in the initial steps of the purification process, critical impurities, including PKA activity, amidolytic activity, and fibrinogen content, are much higher when, for example, the II+III paste is extracted at pH 4.5 or 4.6, as compared to when the extraction occurs at a pH around 4.9 to 5.0 (*see*, Examples 2 to 5).

[0267] In order to counter act the impurities extracted in the methods provided herein, it has now been found that the purity of the IgG composition can be greatly enhanced by the addition of a pretreatment step prior to filtration/centrifugation. In one embodiment, this pretreatment step comprises addition of finely divided silica dioxide particles (*e.g.*, fumed silica, Aerosil®) followed by a 40 to 80 minute incubation period during which the suspension is constantly mixed. In certain embodiments, the incubation period will be between about 50 minutes and about 70 minutes, or about 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, or more minutes. Generally, the treatment will be performed at between about 0°C and about 10°C, or between about 2°C and about 8°C. In certain embodiments, the treatment may be performed at about 0°C, 1°C, 2°C, 3°C, 4°C, 5°C, 6°C, 7°C, 8°C, 9°C, or 10°C. In a particular embodiment, the treatment is performed at between about 2°C and about 10°C.

[0268] The effect of the fumed silica treatment is exemplified by ~~the results found in Example 17. In this example,~~ an experiment where a Fraction II+III precipitate is suspended and split into two samples, one of which is clarified with filter aid only prior to filtration ~~(Figure 7A)~~ and one of which is treated

with fumed silica prior to addition of the filter aid and filtration ~~(Figure 7B)~~. ~~As can be seen in the chromatographs and in the quantitated data,~~ The results of this experiment demonstrate that the filtrate sample pretreated with fumed silica had a much higher IgG purity than the sample only treated with filter aid (68.8% vs. 55.7%~~; compare Tables 17 and 18, respectively~~).

[0269] In certain embodiments, fumed silica is added at a concentration of between about 20 g/kg II+III paste and about 100 g/kg II+III paste (*i.e.*, for a Modified Fraction II+III precipitate that is extracted at a ratio of 1:15, fumed silica should be added at a concentration from about 20 g/16 kg II+III suspension to about 100 g/16 kg II+III suspension, or at a final concentration of about 0.125% (w/w) to about 0.625% (w/w)). In certain embodiments, the fumed silica may be added at a concentration of about 20 g/kg II+III paste, or about 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, or 100 g/kg II+III paste. In one specific embodiment, fumed silica (e.g., Aerosil 380 or equivalent) is added to the Modified Fraction II+III suspension to a final concentration of about 40 g/16 kg II+III. Mixing takes place at about 2 to 8°C for at least 50 to 70 minutes.

[0270] In certain embodiments, SiO_2 is added to a an IgG composition at a concentration between about 0.01 g/g protein and about 10 g/g protein. In another embodiment, SiO_2 is added to a an IgG composition at a concentration between about 0.01 g/g protein and about 5 g/g protein. In another embodiment, SiO_2 is added to an IgG composition at a concentration between about 0.02 g/g protein and about 4 g/g protein. In one embodiment, SiO_2 is added at a final concentration of at least 0.1 g per gram total protein. In another specific embodiment, fumed silica is added at a concentration of at least 0.2 g per gram total protein. In another specific embodiment, fumed silica is added at a concentration of at least 0.25 g per gram total protein. In other specific embodiments, fumed silica is added at a concentration of at least 1 g per gram total protein. In another specific embodiment, fumed silica is added at a concentration of at least 2 g per gram total protein. In another specific embodiment, fumed silica is added at a concentration of at least 2.5 g per gram total protein. In yet other specific embodiments, finely divided silicon dioxide is added at a concentration of at least 0.01 g/g total protein or at least 0.02 g, 0.03 g, 0.04 g, 0.05 g, 0.06 g, 0.07 g, 0.08 g, 0.09 g, 0.1 g, 0.2 g, 0.3 g, 0.4 g, 0.5 g, 0.6 g, 0.7 g, 0.8 g, 0.9 g, 1.0 g, 1.5 g, 2.0 g, 2.5 g, 3.0 g, 3.5 g, 4.0 g, 4.5 g, 5.0 g, 5.5 g, 6.0 g, 6.5 g, 7.0 g, 7.5 g, 8.0 g, 8.5 g, 9.0 g, 9.5 g, 10.0 g, or more per gram total protein.

[0271] In certain embodiments, filter aid, for example Celpure C300 (Celpure) or Hyflo-Supper-Cel (World Minerals), will be added after the silica dioxide treatment, to facilitate depth filtration. Filter aid can be added at a final concentration of from about 0.01 kg/kg II+III paste to about 1.0 kg/kg II+III paste, or from about 0.02 kg/kg II+III paste to about 0.8 kg/kg II+III paste, or from about 0.03 kg/kg II+III paste to about 0.7 kg/kg II+III paste. In other embodiments, filter aid can be added at a final concentration of from about 0.01 kg/kg II+III paste to about 0.07 kg/kg II+III paste, or from about 0.02 kg/kg II+III paste to about 0.06 kg/kg II+III paste, or from about 0.03 kg/kg II+III paste to about 0.05 kg/kg II+III paste. In certain embodiments, the filter aid will be added at a final concentration of about 0.01 kg/kg II+III paste, or about 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, or 1.0 kg/kg II+III paste.

[0272] A significant fraction of IgG was being lost during the filtration step of the GAMMAGARD® LIQUID manufacturing process. It was found that the current methods of post-filtration wash, using 1.8 dead volumes of suspension buffer to purge the filter press frames and lines, were insufficient for maximal recovery of IgG at this step. Surprisingly, it was found that at least 3.0 dead volumes, preferably 3.6 dead volumes, of suspension buffer were required in order for efficient recovery of total IgG in the Modified Fraction II+III clarified suspension (*see*, Example 12 and Figure 1). In certain embodiments, the filter press may be washed with any suitable suspension buffer. In a particular embodiment, the wash buffer will comprise, for example, 5 mM monobasic sodium phosphate, 5 mM acetate, and 0.015% glacial acetic acid (v/v).

[0273] In one aspect, the improvement relates to a method in which a reduced amount of IgG is lost during the Fraction II+III suspension filtration step. In one embodiment, the process improvement is realized by post-washing the filter with at least about 3.6 dead volumes of dissolution buffer containing 150 mL glacial acetic acid per 1000 L. ~~The relationship between the amount of glacial acetic acid and pH in the post-wash buffer is shown in Figure 10.~~ In one embodiment, the pH of the post-wash extraction buffer is between about 4.6 and about 5.3. In a preferred embodiment, the pH of the post-wash buffer is between about 4.7 and about 5.2. In another preferred embodiment, the pH of the post-wash buffer is between about 4.8 and about 5.1. In yet another preferred embodiment, the pH of the post-wash buffer is between about 4.9 and about 5.0.

WHAT IS CLAIMED IS:

1. A method for reducing the amount of a serine protease or a serine protease zymogen in a plasma-derived target protein composition, the method comprising the steps of:

(a) performing a first target protein enrichment step to form a first enriched composition;

(b) contacting the first enriched composition with finely divided silicon dioxide (SiO_2) under conditions suitable to bind at least one serine protease or serine protease zymogen; and

(~~b~~c) separating the SiO_2 from the composition to remove the bound serine protease,

wherein the at least one serine protease or serine protease zymogen is Factor XIa (FXIa), Factor XIIa (FXIIa), Factor XI (FXI), or Factor XII (FXII).

wherein the first target protein enrichment step is a protein precipitation step;

wherein the protein precipitation step is an alcohol fractionation step; and

wherein the first target protein is selected from an immunoglobulin (Ig), albumin, alpha-1-antitrypsin (A1PI), butyrylcholinesterase, a protein of the complement system, and an inter-alpha-trypsin inhibitor (IαI).

~~2. The method of claim 1, wherein the method further comprises the step of performing a first target protein enrichment step to form a first enriched composition, prior to contacting the composition with finely divided silicon dioxide (SiO_2).~~

~~3. The method of claim 2, wherein the first target protein enrichment step is a protein precipitation step.~~

~~4. The method of claim 3, wherein the protein precipitation step is an alcohol fractionation step.~~

~~5. The method of claim 2, wherein the first target protein enrichment step is an ultrafiltration/diafiltration step.~~

~~6~~2. The method of claim 1~~according to any one of claims 2 to 5~~, wherein the method further comprises the step of performing a second target protein enrichment step prior to contacting the enriched composition with finely divided silicon dioxide (SiO_2).

~~7~~3. The method of claim ~~6~~2, wherein the second target protein enrichment step is a protein precipitation step.

~~8~~4. The method of claim ~~7~~3, wherein the protein precipitation step is an alcohol fractionation step.

~~9~~5. The method of claim ~~6~~2, wherein the second target protein enrichment step is an ultrafiltration/diafiltration step.

~~10~~6. The method of claim ~~6~~2, wherein the second target protein enrichment step is a chromatographic enrichment step.

~~11~~7. The method according to any one of claims 1 to ~~10~~6, wherein the method further comprises the step of performing a third target protein enrichment step after contacting the composition with finely divided silicon dioxide (SiO_2).

~~12~~8. The method of claim ~~11~~7, wherein the third target protein enrichment step is a protein precipitation step.

~~13~~9. The method of claim ~~12~~8, wherein the protein precipitation step is an alcohol fractionation step.

~~14~~10. The method of claim ~~12~~8, wherein the third target protein enrichment step is an ultrafiltration/diafiltration step.

~~15~~11. The method of claim ~~12~~8, wherein the third target protein enrichment step is a chromatographic enrichment step.

~~16~~12. The method of claim ~~10 or 15~~6 or 11, wherein the chromatographic enrichment step comprises the sub-steps of:

(i) contacting the plasma-derived target protein composition with a chromatographic resin under conditions suitable to bind the plasma-derived target protein; and

(ii) eluting the plasma-derived target protein from the chromatographic resin.

~~17~~13. The method of claim ~~16~~12, wherein at least one impurity does not bind to the chromatographic resin in sub-step (i).

~~18~~14. The method of claim ~~16~~12, wherein at least one impurity binds to the chromatographic resin in sub-step (i), but is not eluted from the chromatographic resin in sub-step (ii).

~~19~~15. The method of claim ~~10 or 15~~6 or 11, wherein the chromatographic enrichment step comprises the sub-steps of:

(i) contacting the first enriched plasma-derived target protein composition with a chromatographic resin under conditions suitable to bind at least one impurity; and

(ii) separating the resin from the plasma-derived protein composition,

wherein the plasma-derived target protein does not bind to the chromatographic resin in sub-step (i).

~~20~~16. The method according to any one of claims ~~15 to 19~~11, wherein the chromatographic resin is selected from the group consisting of an anion exchange resin, a cation exchange resin, a hydrophobic interaction resin, a mixed mode resin, a hydroxyapatite resin, a ligand affinity resin, an immunoaffinity resin, and a size exclusion resin.

~~21~~17. The method of claim ~~20~~16, wherein the chromatographic resin is an anion exchange resin.

~~22~~18. The method of claim ~~20~~16, wherein the chromatographic resin is a cation exchange resin.

~~23~~19. The method of claim ~~20~~16, wherein the chromatographic resin is a hydrophobic interaction resin.

~~24~~20. The method of claim ~~20~~16, wherein the chromatographic resin is a mixed mode resin.

~~25~~21. The method of claim ~~20~~16, wherein the chromatographic resin is a hydroxyapatite resin.

~~26~~22. The method of claim ~~20~~16, wherein the chromatographic resin is a ligand affinity resin.

~~27~~23. The method of claim ~~20~~16, wherein the chromatographic resin is an immunoaffinity resin.

~~28~~24. The method of claim ~~10 or 15~~6 or 11, wherein the chromatographic enrichment step comprises separating at least one impurity from the target protein by size and/or shape using size exclusion chromatography.

~~29. The method according to any one of claims 1 to 28, wherein the plasma-derived target protein is selected from an immunoglobulin (Ig), albumin, alpha-1-antitrypsin~~

~~(A1PI), butyrylcholinesterase, a protein of the complement system, and an inter-alpha-trypsin inhibitor (IαI).~~

~~30~~25. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is an immunoglobulin (Ig).

~~31~~26. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is albumin.

~~32~~27. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is alpha-1-antitrypsin.

~~33~~28. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is butyrylcholinesterase.

~~34~~29. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is a protein of the complement system.

~~35~~30. The method of claim ~~34~~29, wherein the protein of the complement system is selected from the group consisting of Factor H (FH), Factor D, complement protein C3, and C4 binding protein.

~~36~~31. The method according to any one of claims 1 to 24~~of claim 29~~, wherein the plasma-derived protein is an inter-alpha-trypsin inhibitor.

~~37~~32. The method according to any one of claims 1 to ~~36~~31, wherein the plasma-derived target protein composition is a manufacturing intermediate.

~~38. A method for preparing a plasma-derived Factor H composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:~~

~~(a) contacting a composition containing Factor H and at least one serine protease or serine protease zymogen with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the Factor H and at least one serine protease or serine protease zymogen;~~

~~(b) separating the SiO_2 from the composition;~~

~~(c) eluting the serine protease or serine protease zymogen from the SiO_2 under a solution condition in which a substantial fraction of Factor H remains bound; and~~

(d) eluting the Factor H from the SiO_2.

39. The method of claim 38, wherein the solution condition under which the Factor H and at least one serine protease or serine protease zymogen bind the SiO_2 comprises a pH below 7.0 and a conductivity of less than 11 mS/cm.

40. The method of claim 38 or 39, wherein the solution condition under which the Factor H and at least one serine protease or serine protease zymogen bind the SiO_2 comprises a pH between 4.5 and 6.5 and a conductivity of less than 6 mS/cm.

41. The method of claim 40, wherein the solution condition under which the Factor H and at least one serine protease or serine protease zymogen bind the SiO_2 comprises a pH between 4.5 and 6.5 and a conductivity between 0.5 mS/cm and 5 mS/cm.

42. The method according to any one of claims 38 to 41, wherein the solution condition under which the serine protease or serine protease zymogen is eluted from the SiO_2 and a substantial fraction of the Factor H remains bound comprises a pH below 7.0 and a conductivity of less than 11 mS/cm.

43. The method of claim 42, wherein the solution condition under which the serine protease or serine protease zymogen is eluted from the SiO_2 and a substantial fraction of the Factor H remains bound comprises a pH between 4.5 and 6.5 and a conductivity of less than 6 mS/cm.

44. The method of claim 42, wherein the solution condition under which the serine protease or serine protease zymogen is eluted from the SiO_2 and a substantial fraction of the Factor H remains bound comprises a pH between 5.0 and 6.5 and a conductivity between 0.5 mS/cm and 5 mS/cm.

45. The method according to any one of claims 38 to 44, wherein the solution condition under which the Factor H is eluted from the SiO_2 comprises an ionic strength of at least 6 mS/cm.

46. The method according to any one of claims 38 to 45, wherein the solution condition under which the Factor H is eluted from the SiO_2 comprises an ionic strength of at least 11 mS/cm.

47. The method according to any one of claims 38 to 46, wherein the solution condition under which the Factor H is eluted from the SiO_2 comprises a pH between 5.0 and 7.0.

48. The method according to any one of claims 38 to 44, wherein the solution condition under which the Factor H is eluted from the SiO_2 comprises a pH between 7.0 and 8.0 and an ionic strength of between 4 mS/cm and 7 mS/cm.

49. A method for preparing a Factor H composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a composition containing Factor H and at least one serine protease or serine protease zymogen with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the Factor H and at least one serine protease;

(b) separating the SiO_2 from the composition; and

(c) eluting the Factor H from the SiO_2 under conditions in which a substantial fraction of the serine protease or serine protease zymogen remains bound to the SiO_2.

50. The method of claim 49, wherein the solution condition under which the Factor H and at least one serine protease or serine protease zymogen bind the SiO_2 comprises a pH below 7.0 and a conductivity of less than 11 mS/cm.

51. The method of claim 49 or 50, wherein the solution condition under which the serine protease or serine protease zymogen is eluted from the SiO_2 and a substantial fraction of the Factor H remains bound comprises a pH between 4.5 and 6.5 and a conductivity of less than 6 mS/cm.

52. The method of claim 49, wherein the solution condition suitable to bind the Factor H and at least one serine protease or serine protease zymogen comprises a pH between 5.0 and 6.0 and a conductivity of between 0.5 mS/cm and 5.0 mS/cm.

53. The method according to any one of claims 49 to 52, wherein the solution condition under which the Factor H is eluted from the SiO_2 and a substantial fraction of the serine protease or serine protease zymogen remains bound comprises a pH between 5.0 and 7.0 and a conductivity of at least 11 mS/cm.

54. The method according to any one of claims 49 to 52, wherein the solution condition under which the Factor H is eluted from the SiO_2 and a substantial fraction of the serine protease or serine protease zymogen remains bound comprises a pH between 7.0 and 8.0 and a conductivity of between 2 mS/cm and 10 mS/cm.

55. The method of claim 54, wherein the conductivity of the solution condition is between 4 mS/cm and 7 mS/cm.

56. The method of claim 54, wherein the conductivity of the solution condition is between 5 mS/cm and 6 mS/cm.

57. A method for preparing a Factor H composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a composition containing Factor H and at least one serine protease or serine protease zymogen with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the Factor H but not a substantial fraction of the at least one serine protease or serine protease zymogen;

(b) separating the SiO_2 from the composition; and

(c) eluting the Factor H from the SiO_2.

58. The method of claim 57, wherein the solution condition under which the Factor H binds to the SiO_2 and a substantial fraction of the serine protease or serine protease zymogen does not bind to the SiO_2 comprises a pH of between 5.0 and 7.0 and a conductivity of no more than 14 mS/cm.

59. The method of claim 58, wherein the conductivity of the solution condition is between 9 mS/cm and 14 mS/cm.

60. A method for preparing a Factor H composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a composition containing Factor H and at least one serine protease or serine protease zymogen with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the at least one serine protease or serine protease zymogen but not a substantial fraction of the Factor H; and

(b) separating the SiO_2 from the composition.

61. The method of claim 60, wherein the solution condition under which the serine protease or serine protease zymogen binds to the SiO_2 and a substantial fraction of the Factor H does not bind to the SiO_2 comprises a pH between 5.0 and 7.0 and a conductivity of at least 11 mS/cm.

62. The method of claim 60, wherein the solution condition under which the serine protease or serine protease zymogen binds to the SiO_2 and a substantial fraction of the Factor H does not bind to the SiO_2 comprises a pH between 7.0 and 8.0 and a conductivity of between 2 mS/cm and 10 mS/cm.

63. The method of claim 62, wherein the conductivity of the solution condition is between 4 mS/cm and 7 mS/cm.

64. The method of claim 62, wherein the conductivity of the solution condition is between 5 mS/cm and 6 mS/cm.

65. A method for preparing a Factor H composition, the method comprising the steps of:

(a) contacting a composition containing Factor H and at least one serine protease or serine protease zymogen with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the Factor H;

(b) washing the SiO_2 with a solution comprising a pH between 5.0 and 7.0 and a conductivity of less than 4 mS/cm; and

(c) eluting Factor H from the SiO_2 with a solution comprising a pH between 7.0 and 8.0 and a conductivity greater than 10 mS/cm.

66. The method of claim 65, wherein the solution used to wash the SiO_2 comprises a pH between 5.5 and 6.5.

67. The method of claim 65, wherein the solution used to wash the SiO_2 comprises a pH between of 6.0±0.2.

68. The method according to any one of claims of claims 65 to 67, wherein the solution used to elute Factor H comprises a conductivity of at least 20 mS/cm.

69. The method of claim 68, wherein the solution used to elute Factor H comprises a conductivity of between 25 mS/cm and 40 mS/cm.

70. The method according to any one of claims of claims 38 to 69, wherein the starting composition containing Factor H is a suspended Cohn fraction II+III precipitate, or equivalent fraction thereof.

71. The method according to any one of claims of claims 38 to 69, wherein the starting composition containing Factor H is a suspended Kistler/Nitschmann Precipitate A, or equivalent fraction thereof.

72. The method according to any one of claims of claims 38 to 69, wherein the starting composition containing Factor H is a suspended Kistler/Nitschmann Precipitate B, or equivalent fraction thereof.

73. A method for preparing an inter-alpha-trypsin inhibitor (IαI) composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a solution containing IαI and at least one serine protease with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the IαI and at least one serine protease;

(b) separating the SiO_2 from the composition;

(c) eluting the serine protease or serine protease zymogen from the SiO_2 under conditions in which a substantial fraction of the IαI remains bound; and

(d) eluting the IαI from the SiO_2.

74. A method for preparing an inter-alpha-trypsin inhibitor (IαI) composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a solution containing IαI and at least one serine protease with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the IαI and at least one serine protease;

(b) separating the SiO_2 from the composition; and

(c) eluting the IαI from the SiO_2 under conditions in which a substantial fraction of the serine protease or serine protease zymogen remains bound to the SiO_2.

75. A method for preparing an inter-alpha-trypsin inhibitor (IαI) composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a solution containing IαI and at least one serine protease with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the IαI but not a substantial fraction of the least one serine protease or serine protease zymogen;

(b) separating the SiO_2 from the composition; and

(c) eluting the IαI from the SiO_2.

76. A method for preparing a inter-alpha-trypsin inhibitor (IαI) composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) contacting a solution containing inter-alpha-trypsin inhibitor (IαI) and at least one serine protease with finely divided silicon dioxide (SiO_2) under conditions suitable to bind the serine protease or serine protease zymogen but not the inter-alpha-trypsin inhibitor (IαI); and

(b) separating the SiO_2 from the composition.

77. A method for preparing an Immunoglobulin G (IgG) composition having a reduced amount of a serine protease or a serine protease zymogen, the method comprising the steps of:

(a) precipitating a cryo-poor plasma fraction, in a first precipitation step, with between about 6% and about 10% alcohol at a pH of between about 7.0 and about 7.5 to obtain a first precipitate and a first supernatant;

(b) precipitating IgG from the first supernatant, in a second precipitation step, with between about 20% and about 25% alcohol at a pH of between about 6.7 and about 7.3 to form a second precipitate;

(c) re-suspending the second precipitate to form a suspension;

(d) contacting the suspension with finely divided silicon dioxide (SiO_2) under a solution condition suitable to bind a serine protease or serine protease zymogen; and

(e) separating the SiO_2 from the suspension to form a clarified suspension.

78. The method of claim 77, wherein the method further comprises the steps of:

(f) precipitating IgG from the clarified suspension formed in step (e), in a third precipitation step, with between about 22% and about 28% alcohol at a pH of between about 6.7 and about 7.3 to form a third precipitate;

(g) re-suspending the third precipitate to form a suspension; and

(h) separating the soluble fraction from the suspension formed in step (e), thereby forming an enriched IgG composition.

79. The method of claim 77 or 78, wherein the method further comprises an anion exchange chromatography enrichment step.

80. The method according to any one of claims 77 to 79, wherein the method further comprises a cation exchange chromatography enrichment step.

81. The method according to any one of claims 77 to 80, wherein the method further comprises at least one dedicated viral inactivation or removal step.

82. The method of claim 81, wherein the method comprises a solvent/detergent (S/D) viral inactivation step.

83. The method of claim 81 or 82, wherein the method comprises a nanofiltration step.

84. The method according to any one of claims 81 to 83, wherein the method comprises an incubation step at low pH.

85. The method according to any one of claims 78 to 84, wherein step (b) comprises adjusting the ethanol concentration of the first supernatant formed in step (a) to about 25% (v/v) at a temperature between about -7°C and about -9°C.

86. The method of claim 85, wherein the temperature is about -9°C.

87. The method according to any one of claims 78 to 86, wherein step (c) comprises re-suspending the precipitate of step (b) with a buffer containing phosphate and

~~acetate, wherein the pH of the buffer is adjusted with between 300 mL and 700 mL of glacial acetic acid per 1000 L of buffer.~~

~~88. The method according to any one of claims 78 to 87, wherein step (d) comprises the addition SiO_2 to a final concentration of between about 0.02 grams per gram precipitate formed in step (b) and about 0.06 grams per gram precipitate formed in step (b).~~

~~89. The method according to any one of claims 78 to 88, wherein the solution condition suitable to bind a serine protease or serine protease zymogen comprises a pH between 4.5 and 6.0 and a conductivity of between 0.1 mS/cm and 3 mS/cm.~~

~~90. The method of claim 89, wherein the pH is between 4.9 and 5.3.~~

~~91. The method of claim 89 or 90, wherein the conductivity is between 0.5 mS/cm and 2 mS/cm.~~

~~92. The method according to any one of claims 78 to 91, wherein step (e) comprises the sub-steps of:~~

~~(i) washing the filter press with at least 3 filter press dead volumes of a buffer containing phosphate and acetate, wherein the pH of the buffer is adjusted with between 50 mL and 200 mL of glacial acetic acid per 1000L of buffer, thereby forming a wash solution; and~~

~~(ii) combining the filtrate of step (f) with the wash solution of step (g), thereby forming a solution.~~

~~93. The method of claim 92, further comprising the sub-step of:~~

~~(iii) treating the solution with a detergent.~~

~~94. The method according to any one of claims 78 to 93, wherein step (h) further comprises solvent and detergent (S/D) treatment of the enriched IgG composition.~~

~~95. The method according to any one of claims 78 to 93, wherein the enriched IgG composition obtained in step (h) contains at least 85% of the IgG content found in the cryo-poor plasma fraction used in step (a).~~

~~96. The method of claim 95, wherein the enriched IgG composition obtained in step (h) contains at least 90% of the IgG content found in the cryo-poor plasma fraction used in step (a).~~

~~97. The method according to any one of claims 77 to 96, wherein the amount of a serine protease or a serine protease zymogen has been reduced by at least 90%.~~

~~98. The method of claim 97, wherein the amount of a serine protease or a serine protease zymogen has been reduced by at least 95%.~~

~~99. The method of claim 97, wherein the amount of a serine protease or a serine protease zymogen has been reduced by at least 98%.~~

~~100. The method of claim 97, wherein the amount of a serine protease or a serine protease zymogen has been reduced by at least 99%.~~

~~101. The method according to any one of claims 97 to 100, wherein the serine protease or a serine protease zymogen is FXIa.~~

~~102~~33. The method according to any one of the preceding claims, wherein the composition is contacted with SiO_2 at a final concentration of at least 1 g SiO_2/g protein.

~~103~~34. The method of claim ~~102~~33, wherein the composition is contacted with SiO_2 at a final concentration of at least 2 g SiO_2/g protein.

~~104~~35. The method of claim ~~102~~33, wherein the composition is contacted with SiO_2 at a final concentration of at least 2.5 g SiO_2/g protein.

~~105~~36. The method according to any one of the preceding claims, wherein the serine protease or serine protease zymogen is Factor XI.

~~106~~37. The method according to any one of the preceding claims, wherein the serine protease or serine protease zymogen is Factor XII.

~~107~~38. The method according to any one of the preceding claims, wherein the serine protease or serine protease zymogen is Factor XIa.

39. The method according to any one of the preceding claims, wherein the serine protease or serine protease zymogen is Factor XIIa.

108. The method according to any one of the preceding claims, wherein the serine protease or serine protease zymogen is Factor XIIa.

109. A plasma-derived protein composition prepared by a process comprising a method for reducing serine protease activity according to any one of the preceding claims.

110. The plasma-derived protein composition of claim 109, wherein the composition is formulated for administration to a subject.

111. The plasma-derived protein composition of claim 109, wherein the composition is formulated for intravenous, intramuscular, or subcutaneous administration.

112. The plasma-derived protein composition according to any one of claims 109 to 111, wherein the composition is aqueous.

113. The plasma-derived protein composition according to any one of claims 109 to 111, wherein the composition is lyophilized.

114. Use of a plasma-derived protein composition according to any one of claims 109 to 113 in the preparation of a medicament for treating a disease associated with aberrant activity of a plasma protein in a subject in need thereof.

115. The use of claim 114, wherein the composition comprises a plasma-derived protein is selected from an immunoglobulin (Ig), albumin, alpha-1-antitrypsin (A1PI), butyrylcholinesterase, a protein of the complement system, and an inter-alpha-trypsin inhibitor (IαI).