

We Claim:

1. A process for preparing a highly concentrated antibody composition comprising:
 - a) a first ultrafiltering of a first antibody preparation to provide a second antibody preparation comprising the retentate of the first ultrafiltration;
 - b) a diafiltering of the second antibody preparation to provide a diafiltered intermediate antibody preparation comprising the retentate of the diafiltration; and
 - c) a second ultrafiltering of the diafiltered intermediate antibody preparation to provide a third antibody preparation comprising the retentate of the second ultrafiltration;wherein the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished at 30°C to 70°C and wherein the level of aggregate contaminants in the third antibody preparation is substantially free of aggregates less than 5 weight percent.
2. The process of claim 1, wherein the antibody is an anti-IgE antibody.
3. The process of claim 1 or 2, wherein the antibody is a monoclonal antibody.
4. The process of any one of claims 1 to 3, wherein the antibody is a chimeric antibody, a humanized antibody, or a human antibody.
5. The process of any one of claims 1 to 4, wherein the antibody is an antigen-binding fragment selected from the group consisting of Fab, Fab', F(ab')₂, and Fv fragment.
6. The process of any one of claims 1 to 5, wherein steps b) and c) are carried out at a temperature of 30 °C to ~~about~~ 50°C.
7. The process of any one of claims 1 to 6, wherein steps a), b), and c) are carried out at a temperature of 30 °C to ~~about~~ 50°C.
8. The process of any one of any one of claims 1 to 57, wherein one or more of the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished at from ~~about~~ 35°C to ~~about~~ 50°C.

9. The process of any one of claims 1 to ~~5~~8, wherein one or more of the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished at 45°C plus or minus 5°C.
10. The process of any one of claims 1 to ~~5~~9, wherein one or more of the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished at ~~about~~ 45°C.
11. The process of any one of claims 1 to ~~5~~10, wherein one or more of the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished at from ~~about~~ 40°C to ~~about~~ 50°C.
12. The process of any one of claims 1 to ~~5~~11, wherein the first antibody preparation has an antibody concentration of from ~~about~~ 0.1 to ~~about~~ 10 grams per liter.
13. The process of claim 12, wherein the first antibody preparation has an antibody concentration of ~~about~~ 1 to ~~about~~ 5 grams per liter.
14. The process of any one of claims 1 to ~~5~~13, wherein the second antibody preparation has an antibody concentration of from ~~about~~ 10 to ~~about~~ 50 grams per liter.
15. The process of claim 14, wherein the second antibody preparation has an antibody concentration of ~~about~~ 20 to ~~about~~ 50 grams per liter.
16. The process of claim 15, wherein the second antibody preparation has an antibody concentration of ~~about~~ 20 to ~~about~~ 40 grams per liter.
17. The process of any one of claims 1 to ~~5~~16, wherein the third antibody preparation has an antibody concentration of from ~~about~~ 50 to ~~about~~ 250 grams per liter.
18. The process of claim 17, wherein the third antibody preparation has an antibody concentration of from ~~about~~ 100 to ~~about~~ 230 grams per liter.
19. The process of claim ~~15~~18, wherein the third antibody preparation has an antibody concentration of from ~~about~~ 170 to ~~about~~ 200 grams per liter.

20. The process of any one of claims 1 to 519, wherein the diafiltered intermediate antibody preparation and the third antibody preparation comprise the ultra-filter retentate.
21. The process of any one of claims 1 to 520, wherein the intermediate antibody preparation has an antibody concentration of ~~about~~25 to ~~about~~35 grams per liter and the third antibody preparation has an antibody concentration of from ~~about~~170 to ~~about~~200 grams per liter.
22. The process of claim 21, wherein the first ultrafiltering concentrates the first antibody preparation to provide the second antibody preparation having an antibody concentration of ~~about~~30 grams per liter and the second ultrafiltering concentrates the intermediate antibody preparation to provide the third antibody preparation having an antibody concentration of ~~about~~170 to ~~about~~200 grams per liter.
23. The process of any one of claims 1 to 522, wherein the process is accomplished in from ~~about~~1 to 10 hours.
24. The process of claim 23, wherein the process is accomplished in from ~~about~~2 to 5 hours.
25. The process of claim 24, wherein the process is accomplished in ~~about~~3 hours.
26. The process of any one of claims 1 to 525, wherein the first and the second ultrafiltering are accomplished with an ultra-filter membrane having a nominal pore size of ~~about~~5 to ~~about~~50 kilo Daltons.
27. The process of claim 26, wherein the first and the second ultrafiltering are accomplished with an ultra-filter membrane having a nominal pore size of ~~about~~10 to ~~about~~30 kilo Daltons.
28. The process of any one of claims 1 to 527, wherein the first antibody preparation contains an antibody having an apparent molecular weight of ~~about~~100 to 200 kilo Daltons.

29. The process of claim 28, wherein the first antibody preparation contains an antibody having an apparent molecular weight of ~~about~~ 150 kilo Daltons.
30. The process of any one of claims 1 to 29, wherein the first ultrafiltering concentrates the first antibody preparation to provide the second antibody preparation having an antibody concentration of 30 grams per liter and the second ultrafiltering concentrates the intermediate antibody preparation to provide the third antibody preparation having an antibody concentration of 170 to 200 grams per liter.
31. The process of any one of claims 1 to 530, wherein filtration steps a), b), and c) utilize an ultra-filtration membrane.
32. The process of any one of claims 1 to 531, wherein the first ultrafiltering and the second ultrafiltering are accomplished with the same ultra-filter membrane.
33. The process of any one of claims 1 to 532, wherein an ultra-filtration membrane utilized in filtration steps a) is used in step b) and in step c).
34. The process of any one of claims 1 to 533, wherein the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished with tangential flow filtration across an ultra-filter membrane.
35. The process of claim 34, wherein the first ultrafiltering, the second ultrafiltering, and the diafiltering are accomplished with tangential flow filtration across the same ultra-filter membrane.
36. The process of any one of claims 1 to 535, wherein the first ultrafiltering and the second ultrafiltering are accomplished with a regenerated cellulose composite ultra-filter membrane.
37. The process of any one of claims 1 to 536, wherein the diafiltering accomplishes a buffer exchange at constant volume, constant concentration, or both.
38. The process of any one of claims 1 to 537, wherein the diafiltering accomplishes a buffer exchange of from ~~about~~ 5 to ~~about~~ 15 fold volumes.

39. The process of claim 38, wherein the diafiltering accomplishes a buffer exchange of about 8 fold volumes.
40. ~~The process of any one of claims 1 to 5, wherein step a) comprises more than one diafiltering step.~~
41. ~~The process of claim 40, wherein a first diafiltering step accomplishes a buffer exchange of about 4 fold volumes and a second diafiltering step accomplishes a buffer exchange of about 4 fold volumes.~~
4240. The process of any one of claims 1 to 539, wherein step b) comprises more than one diafiltering step.
4341. The process of claim 4240, wherein a first diafiltering step accomplishes a buffer exchange of about 4 fold volumes and a second diafiltering step accomplishes a buffer exchange of about 4 fold volumes.
4442. The process of any one of claims 1 to 541, wherein the diafiltering exchanges a first buffer for a second buffer.
4543. The process of any one of claims 1 to 542, wherein diafiltering exchanges a buffer having a composition that differs from that of step a).
4644. The process of claim 4543, wherein the a first buffer comprises a mixture of aqueous sodium chloride and a TRIS buffer.
4745. The process of claim 4644, wherein the a second buffer comprises a mixture of aqueous histidine chloride and arginine chloride.
4846. The process of any one of claims 1 to 545, wherein the yield of the third antibody preparation is greater than about 70 weight % based on the weight of antibodies in the first antibody preparation.

~~49~~47. The process of claim ~~48~~46, wherein the yield of the third antibody preparation is from ~~about 80 to about 100~~ weight % based on the weight of antibodies in the first antibody preparation.

~~50~~48. The process of claim ~~49~~47, wherein the yield of the third antibody preparation is greater than ~~about 98~~ weight % based on the weight of antibodies in the first antibody preparation.

~~51~~49. The process of any one of claims 1 to ~~54~~48, wherein the first ultrafiltering has a recirculation rate of from ~~about 0.5 L/min/ft² to about 5 L/min/ft²~~.

~~52~~50. The process of any one of claims 1 to ~~54~~49, wherein the ultrafiltering and diafiltering are accomplished at a transmembrane pressure of from ~~about 5 to about 50 p.s.i.~~

~~53~~51. The process of claim ~~52~~50, wherein the ultrafiltering and diafiltering are accomplished at a transmembrane pressure of from ~~about 10 to about 50 p.s.i.~~

~~54~~52. The process of any one of claims 1 to ~~55~~51, wherein throughput for step c) is at ~~about 80 g/ft²/hr, about 120 g/ft²/hr, about 135 g/ft²/hr, about 145 g/ft²/hr, about 175 g/ft²/hr, about 180 g/ft²/hr, about 265 g/ft²/hr, about 285 g/ft²/hr, or about 290 g/ft²/hr.~~

~~55~~. The process of any one of claims 1 to 5, wherein level of aggregate contaminants is less than 2 weight percent.

~~56~~53. The process of any one of claims 1 to ~~55~~52, wherein there is provided an antibody concentrate with a detectable bio-burden of less than ~~about 100 CFU/mL~~.

~~57~~54. The process of any one of claims 1 to ~~55~~53, wherein the third antibody preparation has a detectable bioburden of 1.8 CFU/ml.

~~58~~55. The process of any one of claims 1 to ~~55~~53, wherein the third antibody preparation has a detectable bioburden of less than 0.13 CFU/ml.

~~59~~56. The process of any one of claims 1 to ~~55~~55, wherein the first antibody preparation has undergone a purification step prior to step a).

~~60~~57. The process of any one of claims 1 to ~~55~~56, wherein the antibody contained in the first antibody preparation is ~~about~~ 99.8% pure prior to step (a).

~~61~~. The process of any one of claims 1 to 5, wherein level of aggregate contaminants in the ~~third antibody preparation is less than 5 weight percent.~~

~~62~~58. The process of any one of claims 1 to ~~55~~57, wherein level of aggregate contaminants in the third antibody preparation is less than 2 weight percent.

~~63~~59. The process of any one of claims 1 to ~~55~~58, wherein the pharmaceutical composition is suitable for therapeutic administration by intramuscular or intravenous administration.

~~64~~60. The process of claim ~~63~~59, wherein the therapeutic administration is to a human.

61. The process of any one of claims 1 to 60, wherein the first antibody preparation is purified antibody preparation.

62. The process of any one of claims 1 to 61, wherein the antibody is anti-IgE antibody rhuMab E25 or an antigen-binding fragment thereof.