PURIFIED ANTIBODY COMPOSITION

RELATED APPLICATIONS

This application claims priority to U.S provisional application Serial No. 60/789,725, filed on April 5, 2006 and to U.S. provisional application Serial No. 60/790,414, filed on April 6, 2006, the contents of each of which are hereby incorporated in their entirety.

BACKGROUND OF THE INVENTION

Large-scale, economic purification of proteins is increasingly an important problem for the biotechnology industry. Generally, proteins are produced by cell culture, using either mammalian or bacterial cell lines engineered to produce the protein of interest by insertion of a recombinant plasmid comprising the gene for that protein. Since the cell lines used are living organisms, they must be fed with a complex growth medium, comprising sugars, amino acids, and growth factors, usually supplied from preparations of animal serum. Separation of the desired protein from the mixture of compounds fed to the cells and from the by-products of the cells themselves to a purity sufficient for use as a human therapeutic poses a formidable challenge.

SUMMARY OF THE INVENTION

There is a need for improved methods of obtaining antibody preparations comprising a reduced amount of host cell protein, including procathepsin L. The invention provides a method for purifying antibodies expressed in a host cell expression system, wherein the resulting antibody preparation comprises a reduced amount of host cell protein, including procathepsin L. The improved method of the invention also includes the the development of reproducible methods of accurately detecting host cell proteins, and a kinetic assay

The invention provides a method for producing a host cell protein-(HCP) reduced antibody preparation from a mixture comprising an antibody and at least one HCP, comprising an ion exchange separation step wherein the mixture is subjected to a first ion exchange material, such that the HCP-reduced antibody preparation is obtained. In one embodiment, the ion exchange separation step comprises passing the mixture over the first ion exchange material such that a first eluate having a reduced level of HCP is obtained. In one embodiment, the method of the invention further comprises a second ion exchange separation step wherein the first eluate is subjected to a second ion exchange material such that a first flowthrough having a reduced level of HCP is obtained. In another embodiment, the method of the invention further comprises a hydrophobic interaction separation step wherein the first flowthrough is subjected to a

What is claimed:

1. A liquid composition comprising adalimumab, wherein the adalimumab is expressed in a Chinese Hamster Ovary (CHO) ceil expression system; and the composition is characterized in that when the composition is assayed in a cathepsin L kinetic assay, a level of cathepsin L activity less than 1.84 RPU/s/mg of adalimumab is observed, wherein the cathepsin L kinetic assay comprises:

i) diluting the composition in a polystyrene container in a solution containing 25 mM NaOAc, 5 mM DTT and 1 mM EDTA at pH 5.5,

ii) adding dextran sulfate to a concentration of 0.035 μg/mL and incubating at 37° C. for six hours,

iii) adding Z-leucine-arginine covalently bound at its C-terminus to a fluorescent 7-amino-4-methyl coumarin (Z-leucine-arginine-AMC), wherein the diluting, adding, and incubating steps are sufficient to permit the measurement of cathepsin L hydrolysis of the Z-leucine-arginine-AMC within a linear range, and

iv) measuring Z-leucine-arginine-AMC hydrolysis in the linear range in RFU/s/mg of adalimumab.

2. The liquid composition of claim 1, wherein the cathepsin L activity is no greater than 1.3 RFU/s/mg of adalimumab.

3. The liquid composition of claim 1, wherein the cathepsin L activity is no greater than 1.0 RFU/s/mg of adalimumab.

4. The liquid composition of claim 1, wherein the cathepsin L activity is no greater than 0.6 RFU/s/mg of adalimumab.

5. The liquid composition of claim 1, wherein the cathepsin L activity is no greater than 0.85 RFU/s/mg of adalimumab.

6. The liquid composition of claim 1, wherein the cathepsin L activity is no greater than 0.9 RFU/s/mg of adalimumab.

7. The liquid composition of claim 1, wherein the composition is packaged in a pre-filled syringe.

8. The liquid composition of claim 1, wherein the composition comprises 50 mg/ml of adalimumab.

9. The liquid composition of claim 1, wherein the composition is suitable for subcutaneous injection.

10. The liquid composition of claim 1, wherein the diluted composition has an adalimumab concentration of 20 µg/ml.

11. The liquid composition of claim 1, wherein the diluted composition has an adalimumab concentration of 50 µg/ml.

12. The liquid composition of claim 1, wherein step i) of the cathepsin L kinetic assay comprises diluting the composition 600 fold.

13. The liquid composition of claim 2, wherein the composition is packaged in a pre-filled syringe.

14. The liquid composition of claim 2, wherein the composition comprises 50 mg/ml of adalimumab.

15. The liquid composition of claim 2, wherein the composition is suitable for subcutaneous injection.

16. The liquid composition of claim 2, wherein the diluted composition has an adalimumab concentration of 20 µg/ml.

17. The liquid composition of claim 2, wherein the diluted composition has an adalimumab concentration of 50 µg/ml.

18. The liquid composition of claim 2, wherein step i) of the cathepsin L kinetic assay comprises diluting the composition 600 fold.

19. The liquid composition of claim 3, wherein the composition is packaged in a pre-filled syringe.

20. The liquid composition of claim 3, wherein the composition comprises 50 mg/ml of adalimumab.

21. The liquid composition of claim 3, wherein the composition is suitable for subcutaneous injection.

22. The liquid composition of claim 3, wherein the diluted composition has an adalimumab concentration of 20 µg/ml.

23. The liquid composition of claim 3, wherein the diluted composition has an adalimumab concentration of 50 µg/ml.

24. The liquid composition of claim 3, wherein step i) of the cathepsin L kinetic assay comprises diluting the composition 600 fold.

25. The liquid composition of claim 4, wherein the composition is packaged in a pre-filled syringe.

26. The liquid composition of claim 4, wherein the composition comprises 50 mg/ml of adalimumab.

27. The liquid composition of claim 4, wherein the composition is suitable for subcutaneous injection.

28. The liquid composition of claim 4, wherein the diluted composition has an adalimumab concentration of 20 µg/ml.

29. The liquid composition of claim 4, wherein the diluted composition has an adalimumab concentration of 50 µg/ml.

30. The liquid composition of claim 4, wherein step i) of the cathepsin L kinetic assay comprises diluting the composition 600 fold.