CLAIMS

1. A method of increasing cell density, viability, and/or titer in a cell culture medium comprising steps of:

(a) providing cells in a cell culture medium to start a cell culture process; and

(b) adding a composition comprising iron to said cell culture medium on or after day 3 or on day 3 and after day 3 of the cell culture process such that the concentration of iron in the cell culture medium is increased over the course of the cell culture process.

2. The method of claim 1, wherein the composition comprising iron is selected from the group consisting of $FeSO_4$, Fe-citrate, Fe-transferrin, Fe-chloride, Fe-nitrate, Fe-EDTA, $Fe(NO_3)_3$, $FeCl_2$, $FeCl_3$ and combinations thereof.

3. The method of claim 1, wherein the composition comprising iron is added on or after day 6 of the cell culture process.

4. The method of claim 1, wherein the composition comprising iron is added at multiple time points during the cell culture process.

5. The method of claim 1, wherein the concentration of iron in the cell culture medium after addition of the composition comprising iron ranges between 100 μM and 5 mM.

6. The method of claim 1, wherein the concentration of iron in the cell culture medium after addition of the composition comprising iron ranges between 300 μM and 1 mM.

7. The method of claim 1, wherein the concentration of iron in the cell culture medium after addition of the composition comprising iron is 1mM.

8. The method of claim 1, wherein the cells are mammalian cells.

9. The method of claim 8, wherein the mammalian cells are selected from BALB/c mouse myeloma line, human retinoblasts (PER.C6), monkey kidney cells, human embryonic kidney line (293), baby hamster kidney cells (BHK), Chinese hamster ovary cells (CHO), mouse sertoli cells, African green monkey kidney cells (VERO-76), human cervical carcinoma cells (HeLa), canine kidney cells, buffalo rat liver cells, human lung cells, human liver cells, mouse mammary tumor cells, TRI cells, MRC 5 cells, FS4 cells, or human hepatoma line (Hep G2).

10. The method of claim 9, wherein the mammalian cells are CHO cells.

11. The method of claim 1, wherein the cell culture process is a fed batch culture process.

12. The method of claim 1, wherein the cell culture process is a batch-refeed culture process.

13. The method of claim 1, wherein the cell culture process is a perfusion culture process.

14. The method of claim 1, wherein the cell culture process is a large-scale production culture process.

15. The method of claim 14, wherein the volume of the cell culture medium is at least about 500 L.

16. The method of claim 1, wherein the cell culture medium does not contain hydrolysates.

17. The method of claim 1, wherein the cell culture medium contains hydrolysates.

18. The method of claim 1, wherein the cells carry a gene that encodes a recombinant protein.

19. The method of claim 18, wherein the recombinant protein is an antibody or fragment thereof.

20. The method of claim 19, wherein the antibody is an anti-IL-22 antibody.

21. The method of claim 19, wherein the antibody is an anti-GDF-8 antibody.

22. The method of claim 19, wherein the antibody is Myo29.

23. The method of claim 19, wherein the antibody is a single domain antibody.

24. The method of claim 23, wherein the single domain antibody is an anti-TNF nanobody.

25. The method of claim 18, wherein the recombinant protein is a glycoprotein.

26. The method of claim 18, wherein the recombinant protein is a therapeutic protein.

27. The method of claim 26, wherein the therapeutic protein is a growth factor, a clotting factor, a cytokine, a fusion protein, a pharmaceutical drug substance, a vaccine, an enzyme, or a Small Modular ImmunoPharmaceutical™ (SMIP).

28. The method of any one of claims 18-27, wherein the method further comprises purifying the recombinant protein.

29. A method of preventing or delaying cell death in a cell culture, the method comprising a step of adding a composition comprising iron at one or more time

points on or after day 3 or on day 3 and after day 3 of the cell culture process.

30. A method of inhibiting apoptosis in a cell culture, the method comprising a step of adding a composition comprising iron at one or more time points subsequent to the beginning of the cell culture process.

31. The method of claim 29 or 30, wherein the composition comprising iron is added on day 6.

32. The method of claim 29 or 30, wherein the iron is added in an amount to effect a concentration in the cell culture medium ranging between 100 μM and 5 mM.

33. The method of claim 29 or 30, wherein the iron is added in an amount to effect a concentration in the cell culture medium ranging between 300 μM and 1 mM.

34. The method of claim 29 or 30, wherein the iron is added in an amount to effect a concentration in the cell culture medium of 1 mM.

35. The method of claim 29 or 30, wherein the cell culture process is a fed batch culture process.

36. The method of claim 29 or 30, wherein the cell culture is a batch-refeed culture.

37. The method of claim 29 or 30, wherein the cell culture is a perfusion culture.

38. The method of claim 29 or 30, wherein the cell culture is a large-scale production culture.

39. The method of claim 38, wherein the volume of the cell culture is at least about 500 L.

40. The method of claim 29 or 30, wherein the composition comprising iron is selected from the group consisting of $FeSO_4$, Fe-citrate, Fe-transferrin, Fe-chloride, Fe-nitrate, Fe-EDTA, $Fe(NO_3)_3$, $FeCl_2$, $FeCl_3$ and combinations thereof.

CLAIMS

1. A method of increasing cell density, viability, and/or titer in a cell culture medium comprising steps of:

(a) providing cells in a cell culture medium to start a cell culture process; and

(b) adding a composition comprising iron to said cell culture medium on or after day 3 of the cell culture process such that the concentration of iron in the cell culture medium is increased over the course of the cell culture process.

2. The method of claim 1, wherein the composition comprising iron is selected from the group consisting of $FeSO_4$, Fe-citrate, Fe-transferrin, Fe-chloride, Fe-nitrate, Fe-EDTA, $Fe(NO_3)_3$, $FeCl_2$, $FeCl_3$ and combinations thereof.

3. The method of claim 1, wherein the composition comprising iron is added on or after day 6 of the cell culture process.

4. The method of claim 1, wherein the composition comprising iron is added at multiple time points during the cell culture process.

5. The method of claim 1, wherein the concentration of iron in the cell culture medium after addition of the composition comprising iron either ranges between 100 μM and 5 mM or 300 μM and 1 mM; or is 1 mM.

6. The method of claim 1, wherein the cells are mammalian cells.

7. The method of claim 6, wherein the mammalian cells are selected from BALB/c mouse myeloma line, human retinoblasts (PER.C6), monkey kidney cells, human embryonic kidney line (293), baby hamster kidney cells (BHK), Chinese hamster ovary cells (CHO), mouse sertoli cells, African green monkey kidney cells (VERO-76), human cervical carcinoma cells (HeLa), canine kidney cells, buffalo rat liver cells, human lung cells, human liver cells, mouse mammary tumor cells, TRI cells,

MRC 5 cells, FS4 cells, or human hepatoma line (Hep G2).

8. The method of claim 7, wherein the mammalian cells are CHO cells.

9. The method of claim 1, wherein the cell culture process is a fed batch culture process, or a batch-refeed culture process, or a perfusion culture process.

10. The method of claim 1, wherein the cell culture process is a large-scale production culture process.

11. The method of claim 10, wherein the volume of the cell culture medium is at least about 500 L.

12. The method of claim 1, wherein the cell culture medium does not contain hydrolysates.

13. The method of claim 1, wherein the cell culture medium contains hydrolysates.

14. The method of claim 1, wherein the cells carry a gene that encodes a recombinant protein.

15. The method of claim 14, wherein the recombinant protein is either:
an antibody or fragment thereof; or
a glycoprotein or a therapeutic protein.

16. The method of claim 15, wherein the antibody is an anti-IL-22 antibody, or an anti-GDF-8 antibody, or the antibody is Myo29, or the antibody is a single domain antibody.

17. The method of claim 16, wherein the single domain antibody is an anti-TNF nanobody.

18. The method of claim 15, wherein the therapeutic protein is a growth factor, a clotting factor, a cytokine, a fusion protein, a pharmaceutical drug substance, a vaccine, an enzyme, or a Small Modular ImmunoPharmaceutical™ (SMIP).

19. The method of any one of claims 14-18, wherein the method further comprises purifying the recombinant protein.

20. A method of preventing or delaying cell death in a cell culture, the method comprising a step of adding a composition comprising iron at one or more time points on or after day 3 of the cell culture process.

21. A method of inhibiting apoptosis in a cell culture, the method comprising a step of adding a composition comprising iron at one or more time points subsequent to the beginning of the cell culture process.

22. The method of claim 20 or claim 21, wherein the composition comprising iron is added on day 6.

23. The method of claim 20 or claim 21, wherein the iron is either added in an amount to effect a concentration in the cell culture medium ranging between 100 μM and 5 mM, or 300 μM and 1 mM; or is added in an amount to effect a concentration in the cell culture medium of 1 mM.

24. The method of claim 20 or claim 21, wherein the cell culture process is a fed batch culture process, or a batch-refeed culture, or a perfusion culture.

25. The method of claim 20 or claim 21, wherein the cell culture is a large-scale production culture.

26. The method of claim 25, wherein the volume of the cell culture is at least about 500 L.

27. The method of claim 20 or claim 21, wherein the composition comprising iron is selected from the group consisting of $FeSO_4$, Fe-citrate, Fe-transferrin, Fe-chloride, Fe-nitrate, Fe-EDTA, $Fe(NO_3)_3$, $FeCl_2$, $FeCl_3$ and combinations thereof.