

CLAIMS:

1. A method for manufacturing a protein of interest wherein said protein is expressed in a host cell, comprising:
 - a) culturing said host cells under conditions such that they express said protein,
 - b) collecting the host cells from the cell culture fluid,
 - c) adding buffer to said host cells, and
 - d) subjecting the host cells to heat treatment wherein the redox potential of said buffer is maintained below -0 mV during said heat treatment.
2. A method according to claim 1 wherein the heat treatment is performed in a container and said redox potential is maintained by reducing the amount of oxygen (O₂) present in the gaseous phase in the container during protein extraction.
3. A method according to claim 1 or 2, wherein nitrogen (N₂) is added to the gaseous phase of the container.
4. A method according to claim 3 wherein said gaseous phase contains at least 50% N₂.
5. A method according to claim 3 or 4 wherein said N₂ is added to the container as an overlay in the gaseous phase of the container or by sparging the N₂ through the buffer containing the host cells obtained in c).
6. A method according to any preceding claim wherein said heat treatment step is performed at 30°C to 70°C.
7. A method according to claim 5 wherein said heat treatment step is performed at 55°C to 65°C.
8. A method according to any preceding claim wherein said temperature is maintained for 1 to 18 hours.
9. A method according to any preceding claim wherein pH of buffer following addition to the host cells and prior to the heat treatment is measured.
10. A method according to claim 9 wherein the pH of the buffer is or is adjusted to a pH of 6 to 9.
11. A method according to any preceding claim wherein the protein is a recombinant protein encoded in an expression vector.
12. A method according to claim 11 wherein the protein is a recombinant antibody.
13. A method according to claim 12, wherein said recombinant antibody is a recombinant antibody fragment.

14. A method according to claim 12 or 13, wherein said recombinant antibody or recombinant antibody fragment specifically binds to TNF-alpha or CD154.