

## WHAT IS CLAIMED IS :

1. A method for selectively reducing CysL97 in a preparation of IL-17 antibodies that have been recombinantly produced by mammalian cells, comprising:
  - a) contacting the preparation with at least one reducing agent in a system to form a reducing mixture; and
  - b) incubating the reducing mixture while maintaining a volumetric oxygen mass-transfer coefficient ( $k_{La}^*$ ) in the system of  $\leq$  about  $0.37 \text{ h}^{-1}$ , said  $k_{La}^*$  being calculated by adapting a dissolved oxygen curve to a saturation curve;wherein the IL-17 antibodies each comprise an immunoglobulin heavy chain variable domain ( $V_H$ ) comprising the three complementarity determining regions (CDRs) of the  $V_H$  set forth as SEQ ID NO:8 and an immunoglobulin light chain variable domain ( $V_L$ ) comprising the three CDRs of the  $V_L$  set forth as SEQ ID NO:10, and further wherein prior to step a) the initial percent oxygen saturation in the preparation is at least about 60%, as measured using an oxygen probe calibrated at  $25^\circ\text{C}$ .
2. The method according to claim 1, wherein the at least one reducing agent is a thiol-containing reducing agent having a standard oxidation-reduction potential,  $E^0$ , of about  $-0.20 \text{ V}$  to about  $-0.23 \text{ V}$  at pH 7.0, as measured by thiol-disulfide exchange.
3. The method according to claim 1, wherein the at least one reducing agent is cysteine.
4. The method according to claim 3, wherein the  $k_{La}^*$  in the system during step b) is:
  - a.  $\leq$  about  $0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is between about 56:1 to about 118:1, and wherein the reducing mixture is incubated according to step b) for up to about 240 minutes;
  - b.  $\leq$  about  $0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is between about 77:1 to about 118:1, and wherein the reducing mixture is incubated according to step b) for up to about 300 minutes.
  - c.  $<$  about  $0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is between about 46:1 to about 118:1;

- d.  $< \text{about } 0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is between about 54:1 to about 82:1; or
  - e.  $\leq \text{about } 0.27 \text{ h}^{-1}$ .
- 5. The method according to claim 4, wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is about 66:1.
  - 6. The method according to claim 3, wherein the concentration of cysteine in the reducing mixture is about 4.0 mM to about 8.0 mM.
  - 7. The method according to any one of the preceding claims, wherein between step a) and step b), the reducing mixture is heated to a temperature between about 32°C to about 42°C for about 45 to about 90 minutes, and wherein the  $k_{La}^*$  in the system during heating is  $\leq \text{about } 0.69 \text{ h}^{-1}$ , said  $k_{La}^*$  being calculated by adapting a saturation curve to a dissolved oxygen curve.
  - 8. The method according to any one of the preceding claims, wherein the reducing mixture is incubated according to step b) at a temperature between about 20°C to about 42°C for about 210 to about 420 minutes.
  - 9. The method according to any one of the above claims, wherein an oxidized form of the reducing reagent is not added to the reducing mixture.
  - 10. The method according to any one of the above claims, wherein a denaturant is not added to the reducing mixture.
  - 11. A method for selectively reducing CysL97 in a secukinumab preparation that has been recombinantly produced by mammalian cells, comprising:
    - a) contacting the preparation with about 4.0 mM to about 8.0 mM in a system to form a reducing mixture;
    - b) heating the reducing mixture to a temperature between about 32°C to about

42°C for about 45 to about 90 minutes, wherein the volumetric oxygen mass-transfer coefficient ( $k_{La}^*$ ) in the system during heating is  $< \text{about } 0.69 \text{ h}^{-1}$ , said  $k_{La}^*$  being calculated by adapting a saturation curve to a dissolved oxygen curve; and

- c) incubating the reducing mixture at a temperature between about 20°C to about 42°C while maintaining the  $k_{La}^*$  in the system:
- i.  $\leq \text{about } 0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:secukinumab in the reducing mixture is between about 56:1 to about 118:1, and wherein the reducing mixture is incubated for up to about 240 minutes;
  - ii.  $\leq \text{about } 0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:secukinumab in the reducing mixture is between about 77:1 to about 118:1, and wherein the reducing mixture is incubated for up to about 300 minutes;
  - iii.  $< \text{about } 0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:secukinumab in the reducing mixture is between about 46:1 to about 118:1;
  - iv.  $< \text{about } 0.37 \text{ h}^{-1}$ , and wherein the molar ratio of cysteine:secukinumab in the reducing mixture is between about 54:1 to about 82:1; or
  - v.  $\leq \text{about } 0.27 \text{ h}^{-1}$ ,

said  $k_{La}^*$  being calculated by adapting a dissolved oxygen curve to a saturation curve, wherein prior to step a) the initial percent oxygen saturation in the preparation is at least about 60%, as measured using an oxygen probe calibrated at 25°C.

12. A method for selectively reducing CysL97 in a preparation of IL-17 antibodies that have been recombinantly produced by mammalian cells, comprising:

a) contacting the preparation with a set of oxidation/reduction reagents selected from cysteine/cystine and cysteine/cystamine to form a reducing mixture; and

b) incubating the reducing mixture at a temperature of:

- i. about 37 °C under anaerobic conditions for at least about 4 hours,  
or
- ii. about 18-24 °C for about 16-24 hours;

wherein the IL-17 antibodies each comprise an immunoglobulin heavy chain variable domain ( $V_H$ ) comprising the three complementarity determining regions (CDRs)

of the V<sub>H</sub> set forth as SEQ ID NO:8 and an immunoglobulin light chain variable domain (V<sub>L</sub>) comprising the three CDRs of the V<sub>L</sub> set forth as SEQ ID NO:10.

13. The method according to claim 12, wherein the molar ratio of oxidation/reduction reagents in the reducing mixture is between about 4:1 to about 80:1.

14. The method according to claim 12, wherein the set of oxidation/reduction reagents is cysteine/cystine or cysteine/cystamine.

15. The method according to claim 14, wherein the molar ratio of cysteine:IL-17 antibodies in the reducing mixture is between about 21:1 to about 296:1.

16. The method according to any one of the above claims, wherein:

- i. the three CDRs of the V<sub>H</sub> set forth as SEQ ID NO:8 are, in sequence, the amino acids sequences set forth as SEQ ID NOs: 1, 2, and 3 or the amino acids sequences set forth as SEQ ID NOs: 11, 12, and 13, and the three CDRs of the V<sub>L</sub> set forth as SEQ ID NO:10 are, in sequence, the amino acids sequences set forth as SEQ ID NOs: 4, 5 and 6;
- ii. the IL-17 antibodies each comprise the V<sub>L</sub> set forth as SEQ ID NO:10 and the V<sub>H</sub> set forth as SEQ ID NO:8; or
- iii. the IL-17 antibodies each comprise the full length light chain set forth as SEQ ID NO:14 and the full length heavy chain set forth as SEQ ID NO:15, with or without the C-terminal lysine.

17. The method according to any one of the above claims, wherein the IL-17 antibodies are human antibodies of the IgG<sub>1</sub> isotype.

18. The method according to any one of the above claims, wherein the IL-17 antibodies are secukinumab antibodies.

19. The method according to any one of the above claims, wherein the level of activity of the IL-17 antibodies in the preparation increases by at least about 10 percentage points within about 60 minutes following step b), as measured by cystamine-CEX.

20. A method for selectively reducing CysL97 in a preparation of secukinumab antibodies that have been recombinantly produced by mammalian cells, comprising:

- a) adjusting the concentration of secukinumab in the preparation to between about 4 mg/ml – about 19.4 mg/ml;
- b) adjusting the percent oxygen saturation in the preparation to at least about 60%;
- c) adjusting the pH of the preparation to about 7.4 - about 8.5;
- d) contacting the preparation with cysteine in a vessel to form a reducing mixture, wherein the concentration of cysteine in the reducing mixture is about 4.0 mM - about 8.0 mM;
- e) heating the reducing mixture to a temperature between about 32°C - about 42°C;
- f) incubating the reducing mixture from step e) at a temperature between about 20°C - about 42°C, said incubating occurring for about 210 - about 420 minutes, while maintaining a volumetric oxygen mass-transfer coefficient ( $k_{La}^*$ ) in the vessel of  $\leq 0.37 \text{ h}^{-1}$ , said  $k_{La}^*$  being calculated by adapting a saturation curve to a dissolved oxygen curve,
- g) cooling the mixture resultant from step f) to a temperature between about 16°C - about 28°C, said cooling occurring for about 45 - about 90 minutes; and
- h) adjusting the pH of the mixture resultant from step g) to between about 5.1 - about 5.3.

21. A purified preparation of IL-17 antibodies produced by the method set forth any one of the preceding claims.

22. A purified preparation of secukinumab, wherein the level of intact secukinumab in the preparation is at least about 90%, as measured by sodium dodecyl sulfate capillary electrophoresis (CE-SDS), and wherein the level of activity of secukinumab in the preparation is at least about 90%, as measured by cystamine-CEX.