

Claims

1. A method for the purification of an antibody by chromatography, comprising the following steps:
- 5
- a) loading a sample comprising an antibody onto a chromatography resin,
 - b) optionally, washing the resin,
 - c) applying an elution buffer to the chromatography resin, and
 - d) starting collection of the eluate,
- 10
- wherein the collection of the eluate is started at a predetermined interval (D0) after the absorbance signal of the eluate has reached a predetermined value (A0), and wherein the predetermined value (A0) is in the range of 10 - 50% of the absorbance signal at peak maximum of elution peaks obtained by different elution runs with the antibody sample to be purified.
- 15
2. The method according to claim 1, wherein the different elution runs vary in pH, load density or salt conditions.
- 20
3. The method according to claim 2, wherein the varying conditions are within a range of pH 5 to pH 7 and within a range of 5 to 50 g/L resin load.
- 25
4. The method according to claim 3, wherein the predetermined absorbance value (A0) is in the range of 0 to 1500 mAU measured at 280 nm.
- 30
5. The method according to claim 4, wherein the predetermined absorbance value (A0) is 700 mAU measured at 280 nm.
- 35
6. The method according to any of the preceding claims, wherein the predetermined interval (D0) is a flow volume interval that is determined by the following steps:
- a) receiving the elution peak chromatograms of at least two different elution runs (ER₁, ..., ER_N) with the antibody sample to be purified, wherein the different elution runs vary in pH, load density or salt conditions,
 - b) specifying an absorbance signal A₁, ..., A_N for each of the elution peaks received in step a), wherein each of the corresponding eluate fractions of A₁, ..., A_N has an aggregate/impurity content below 4%,

- 5 c) determining the flow volumes $C_1, \dots, C_N$ of the absorbance signals $A_1, \dots, A_N$ for each of the elution runs in the chromatograms,
- d) calculating the difference for each of said flow volumes $C_1, \dots, C_N$, to the flow volume C_0 of A_0 , and
- 5 e) averaging said differences (i.e. obtaining the mean value), thereby obtaining the predetermined flow volume interval (D0).
7. The method according claim 6, wherein the distance of $A_1, \dots, A_N$ to the absorbance signal A_{MAX} at height maximum of the corresponding elution peak is in the range of 0 to 300 mAU
- 10 measured at 280 nm.
8. The method according to claim 7, wherein the predetermined interval value (D0) is between 0.4 to 1.2 CV flow volumes.
- 15 9. The method according claim 8, wherein the predetermined interval value (D0) is 0.6 CV flow volumes.
10. The method according to any of the preceding claims, wherein the chromatography is ion exchange (IEX) chromatography.
- 20 11. The method according to claim 10, wherein the chromatography is cation exchange (CEX) chromatography.
12. The method according to any of the preceding claims, wherein the monoclonal antibody comprises a VH of SEQ ID NO: 8 and a VL of SEQ ID NO: 7.
- 25 13. The method according to claim 12, wherein the antibody comprises a heavy chain of SEQ ID NO: 10 and a light chain of SEQ ID NO: 9.
- 30 14. A pharmaceutical composition obtained by the method according to any of preceding the claims.