

ENGINEERING MONOCLONAL ANTIBODIES TO IMPROVE STABILITY AND PRODUCTION TITER

RELATED APPLICATIONS

5 The present application claims the benefit of U.S. Provisional App. No. 62/787,867 filed January 3, 2019, which is incorporated by reference in its entirety herein.

FIELD

10 The presented subject matter relates to the field of protein engineering. Specifically, the presented subject matter relates to engineering antibodies, especially monoclonal antibodies, and variants thereof, to improve their stability and production.

BACKGROUND

15 Monoclonal antibodies (mAbs) that are recombinantly produced (and active fragments thereof) are important therapeutic tools. However, as these molecules are complex, many challenges need to be met to facilitate production, storage, and therapeutic administration of these molecules.

Two challenges concern production and stability. mAbs are produced in bioreactors from engineered cells, such as Chinese Hamster Ovary (CHO) cells. However, production levels can be low and can vary between mAbs. Low production levels increase production costs, including production time, labor, and consumed resources, such as the necessary components for operating the bioreactor.
20 Furthermore, a lack of stability impacts the “shelf-life” of mAbs. Degraded mAbs can be less potent, and fragmented mAbs can present an immunologic risk.

Therefore there is a need to improve mAb stability and production titer.

SUMMARY

25 In one aspect, the present disclosure provides a method of increasing stability of a first antibody, comprising substituting a hydrophobic amino acid residue at heavy chain position 80 according to AHo numbering of the first antibody to create a second antibody, wherein the second antibody is more stable than the unsubstituted first antibody, wherein the hydrophobic amino acid residue is selected from the group consisting of: alanine, phenylalanine, leucine, methionine, and valine.
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CLAIMS

1. A method of increasing stability of a first antibody comprising substituting a hydrophobic amino acid residue at heavy chain position 80 according to AHo numbering of the first antibody to create a second antibody, wherein the second antibody is more stable than the unsubstituted first antibody, wherein the hydrophobic amino acid residue is selected from the group consisting of: alanine, phenylalanine, leucine, methionine, and valine.
2. The method of claim 1, wherein the hydrophobic amino acid residue is selected from the group consisting of: phenylalanine, leucine, and valine.
3. The method of claim 1, wherein the methionine or isoleucine is substituted at heavy chain position 80 according to AHo numbering of the first antibody.
4. The method of any of claims 1-3, wherein the increased stability of the second antibody is demonstrated by at least one selected from the group consisting of an increase in titer during cell culture, a reduction in high molecular weight species, and an increased melting point temperature.
5. The method of any preceding claim, wherein the first antibody is a monoclonal antibody.
6. The method of any preceding claim, wherein the first antibody is a human monoclonal antibody, a chimeric antibody or a humanized monoclonal antibody.
7. The method of any preceding claim, wherein the first antibody is an IgG antibody selected from the group consisting of an IgG1, IgG2, IgG3, and IgG4 antibody.
8. The method of any of claims 1-7, wherein the first antibody is a multi-specific antibody.
9. The method of claim 8, wherein the multi-specific antibody is a bispecific antibody or trispecific antibody.

10. The method of any of claims 1-9, further comprising formulating the second antibody into a pharmaceutical composition.