ABETA ANTIBODY FORMULATION

The present invention relates to a pharmaceutical formulation of an antibody molecule, and/or a mixture of antibody molecules against the amyloid-beta peptide (Abeta).

Antibody molecules, as part of the group of protein pharmaceuticals, are very susceptible to physical and chemical degradation, such as denaturation and aggregation, deamidation, oxidation and hydrolysis. Protein stability is influenced by the characteristics of the protein itself, e.g. the amino acid sequence, and by external influences, such as temperature, solvent pH, excipients, interfaces, or shear rates. So, it is important to define the optimal formulation conditions to protect the protein against degradation reactions during manufacturing, storage and administration. (Manning, M. C., K. Patel, et al. (1989). "Stability of protein pharmaceuticals." Pharm Res 6(11): 903-18., Zheng, J. Y. and L. J. Janis (2005). "Influence of pH, buffer species, and storage temperature on physicochemical stability of a humanized monoclonal antibody LA298." Int)_Pharm.). Administration of antibodies via subcutaneous or intramuscular route requires high protein concentration in the final formulation due to the often required high doses and the limited administration volumes. (Shire, S. J., Z. Shahrokh, et al. (2004). "Challenges in the development of high protein concentration formulations." J Pharm Sci 93(6): 1390-402., Roskos, L. K., C. G. Davis, et al. (2004). "The clinical pharmacology of therapeutic monoclonal antibodies." Drug Development Research 61(3): 108-120.) The large-scale manufacturing of high protein concentration can be achieved by ultrafiltration processes, drying process, such as lyophilisation or spray-drying, and precipitation processes. (Shire, S.1., Z. Shahrokh, et al. (2004). "Challenges in the development of high protein concentration formulations." J Pharm Sci 93(6): 1390-402.).

It is an object of the ~~present invention~~ embodiments disclosed herein ~~is~~ to provide a highly concentrated, stable formulation of an Abeta antibody or of mixtures of such antibodies, which allows subcutaneous administration of the antibody to a patient.

The formulation of the present ~~invention~~ disclosure shows good stability upon storage for 8 months at 2-8°C and 25°C without formation of visible particles. Shaking and multiple freezing-thawing steps were applied to the liquid formulation to simulate physical stress conditions that potentially occur during manufacturing or transportation of the drug product.

The pharmaceutical formulation of the present disclosure ~~invention~~ comprises a poloxamer as surfactant to reduce aggregation of the antibodies and particle formation. The term "poloxamer" as used herein includes a polyoxyethylene-polyoxypropylene triblock copolymer known as poloxamer 188, sold under the trade name PLURONIC® F68 by BASF (Parsippany, N.J.). Other poloxamers which may be utilized in the formulations of the present disclosure ~~invention~~ include poloxamer 403 (sold as PLURONIC® P123), poloxamer 407 (sold as PLURONIC® P127), poloxamer 402 (sold as PLURONIC® P122), poloxamer 181 (sold as PLURONIC® L61), poloxamer 401 (sold as PLURONIC® L121), poloxamer 185 (sold as PLURONIC® P65), and poloxamer 338 (sold as PLURONIC® F108).

The present disclosure ~~invention~~ provides a stable liquid pharmaceutical antibody formulation comprising:

- about 50 mg/ml – 200 mg/ml of an Abeta antibody,
- about 0.01 % – 0.1% of a poloxamer, preferably poloxamer 188,
- about 5 mM – 50 mM of a buffer,
- about 100 mM – 300 mM of a stabilizer,

at a pH of about 4.5 – 7.0.

In a first aspect, the present disclosure provides a stable liquid pharmaceutical antibody formulation comprising:

- 50 mg/ml – 200 mg/ml of an Abeta antibody,
- 0.01 % – 0.1% of a poloxamer, preferably poloxamer 188,
- 5 mM – 50 mM of a buffer,
- 100 mM – 300 mM of a stabilizer,

at a pH of 4.5 – 7.0,
wherein the Abeta antibody is a monoclonal antibody comprising a heavy chain and a light chain, wherein the heavy chain of the Abeta antibody comprises a VH domain which comprises:

- a CDR1 comprising the amino acid sequence of Seq. Id. No. 4,
- a CDR2 comprising the amino acid sequence of Seq. Id. No. 5,
- a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 6, and

the light chain of the Abeta antibody comprises a VL domain which comprises:

– a CDR1 comprising the amino acid sequence of Seq. Id. No. 7,

– a CDR2 comprising the amino acid sequence of Seq. Id. No. 8,

a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 9

In a particular embodiment of the present ~~invention~~disclosure, the Abeta antibody concentration is about 100 mg/ml – 200 mg/ml, preferably about 150 mg/ml.

In a particular embodiment of the present disclosure~~invention~~, the poloxamer is present in a concentration of about 0.02% – 0.06%, preferably about 0.04 %.

In a particular embodiment of the present disclosure~~invention~~, the buffer is a sodium acetate buffer or a Histidine buffer, preferably a Histidine/Histidine-HCl buffer.

In a particular embodiment of the present disclosure~~invention~~, the buffer has a concentration of about 10 to 30 mM, preferably about 20 mM.

In a particular embodiment of the present disclosure~~invention~~, the pH of the formulation is about 5 – 6, preferably about 5.5.

In a particular embodiment of the present disclosure~~invention~~, the stabilizer is selected from sugars and amino acids.

In a particular embodiment of the present disclosure~~invention~~, the stabilizer is selected from trehalose and arginine.

In a particular embodiment of the present disclosure~~invention~~, the stabilizer has a concentration of about 100 mM to 300 mM.

In a particular embodiment of the present disclosure~~invention~~, the stabilizer is threhalose and has a concentration of about 150 mM to 250 mM, preferably about 200 mM.

In a particular embodiment of the present disclosure~~invention~~, the stabilizer is arginine and has a concentration of about 100 mM to 150 mM, preferably about 135 mM.

In a particular embodiment of the present disclosure~~invention~~, the Abeta antibody is a monoclonal antibody comprising a heavy chain and a light chain.

In a particular embodiment of the present disclosure~~invention~~, the heavy chain of the Abeta antibody comprises a VH domain which comprises:

- a CDR1 comprising the amino acid sequence of Seq. Id. No. 4,
- a CDR2 comprising the amino acid sequence of Seq. Id. No. 5,
- a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 6.

In a particular embodiment of the present disclosure ~~invention~~ the light chain of the Abeta antibody comprises a VL domain which comprises:

- a CDR1 comprising the amino acid sequence of Seq. Id. No. 7,
- a CDR2 comprising the amino acid sequence of Seq. Id. No. 8,
- a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 9.

In a particular embodiment of the present disclosure~~invention~~, the VH domain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 2 and the VL domain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 3.

In a particular embodiment of the present disclosure~~invention~~, the heavy chain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 10.

In a particular embodiment of the present disclosure~~invention~~, the light chain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 11.

In a particular embodiment of the present disclosure~~invention~~, the monoclonal Abeta antibody is a mixture of mono-glycosylated Abeta antibodies and double-glycosylated Abeta antibodies, wherein the mono-glycosylated antibody comprises a glycosylated asparagine (Asn) at position 52 of Seq. Id. No. 2 in the VH domain of one antibody binding site and wherein the double-glycosylated antibody comprises a glycosylated asparagine (Asn) at position 52 of Seq. Id. No. 2 in the VH domain of both antibody binding sites and whereby said mixture comprises less than 5% of an antibody being non-glycosylated at position 52 of Seq. Id. No. 2 in the VH domain.

In a particular embodiment the present disclosure ~~invention~~ provides the use of the pharmaceutical formulation of the present disclosure ~~invention~~ for the subcutaneous administration of the Abeta antibody.

In a second aspect, the present disclosure provides for the pharmaceutical formulation as defined herein for use in the subcutaneous administration of the Abeta antibody.

The terms "Abeta antibody" and "an antibody that binds to Abeta" refer to an antibody that is capable of binding Aβ peptide with sufficient affinity such that the antibody is useful as a diagnostic and/or therapeutic agent in targeting Aβ peptide.

It is of note that Aβ has several naturally occurring forms, whereby the human forms are referred to as the above mentioned Aβ39, Aβ40, Aβ41, Aβ42 and Aβ43. The most prominent form, Aβ42, has the amino acid sequence (starting from the N-terminus):

DAEFRHDSGYEVHHQKLVFFAEDVGSNKGAIIGLMVGGVVIA (Seq. Id. No. 1). In Aβ41, Aβ 40, Aβ 39, the C-terminal amino acids A, IA and VIA are missing, respectively. In the Aβ 43 form an additional threonine residue is comprised at the C-terminus of the above depicted sequence (Seq. Id. No. 1).

The term "mono-glycosylated Abeta antibody" relates to an antibody molecule comprising an N-glycosylation at position 52 of Seq. Id. No. 2 in one (VH)-region of an individual antibody molecule; see also figure 1. The term "double-glycosylation Abeta antibody" defines an antibody molecule which is N-glycosylated at position 52 of Seq. Id. No. 2 on both variable regions of the heavy chain" (figure 1). Antibody molecules which lack a N-glycosylation on both heavy chain (VH)-domains are named "non-glycosylated antibodies" (figure 1). The mono-glycosylated antibody, the double-glycosylated antibody and the non-glycosylated antibody may comprise the identical amino acid sequences or different amino acid sequences. The mono-glycosylated antibody and the double-glycosylated antibody are herein referred to as "glycosylated antibody isoforms". A purified antibody molecule characterized in that at least one antigen binding site comprises a glycosylation in the variable region of the heavy chain (VH) is a mono-glycosylated antibody which is free of or to a very low extent associated with an isoform selected from a double-glycosylated antibody and a nonglycosylated antibody, i.e. a "purified mono-glycosylated antibody". A double-glycosylated antibody in context of this invention is free of or to a very low extent associated with an isoform selected from a mono-glycosylated antibody and a non-glycosylated antibody, i.e. a "purified double-glycosylated antibody".

Claims

1. A stable liquid pharmaceutical antibody formulation comprising:

- ~~about~~ 50 mg/ml – 200 mg/ml of an Abeta antibody,
- ~~about~~ 0.01 % – 0.1% of a poloxamer, preferably poloxamer 188,
- ~~about~~ 5 mM – 50 mM of a buffer,
- ~~about~~ 100 mM – 300 mM of a stabilizer,

at a pH of ~~about~~ 4.5 – 7.0,
wherein the Abeta antibody is a monoclonal antibody comprising a heavy chain and a light chain, wherein the heavy chain of the Abeta antibody comprises a VH domain which comprises:

- a CDR1 comprising the amino acid sequence of Seq. Id. No. 4,
- a CDR2 comprising the amino acid sequence of Seq. Id. No. 5,
- a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 6, and

the light chain of the Abeta antibody comprises a VL domain which comprises:

- a CDR1 comprising the amino acid sequence of Seq. Id. No. 7,
- a CDR2 comprising the amino acid sequence of Seq. Id. No. 8,

a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 9.

2. The pharmaceutical formulation of claim 1, wherein the Abeta antibody concentration is ~~about~~ 100 mg/ml – 200 mg/ml, preferably ~~about~~ 150 mg/ml.

3. The pharmaceutical formulation of claim 1 or 2, wherein the poloxamer is present in a concentration of ~~about~~ 0.02% – 0.06%, preferably ~~about~~ 0.04 %.

4. The pharmaceutical formulation of claims 1 to 3, wherein the buffer is a sodium acetate buffer or a Histidine buffer, preferably a Histidine/Histidine-HCl buffer.

5. The pharmaceutical formulation of claims 1 to 4, wherein the buffer has a concentration of ~~about~~ 10 to 30 mM, preferably ~~about~~ 20 mM.

6. The pharmaceutical formulation of claims 1 to 5, wherein the pH of the formulation is ~~about~~ 5 – 6, preferably ~~about~~ 5.5.

7. The pharmaceutical formulation of claims 1 to 6, wherein the stabilizer is selected from sugars and amino acids.

8. The pharmaceutical formulation of claim 7, wherein the stabilizer is selected from trehalose and arginine.

9. The pharmaceutical formulation of claim 7 or 8, wherein the stabilizer has a concentration of ~~about~~ 100 mM to 300 mM.

10. The pharmaceutical formulation of claim 8 or 9, wherein the stabilizer is ~~th~~rehalose and has a concentration of ~~about~~ 150 mM to 250 mM, preferably ~~about~~ 200 mM.

~~10~~11. The pharmaceutical formulation of claim 8 or 9, wherein the stabilizer is arginine and has a concentration of ~~about~~ 100 mM to 150 mM, preferably ~~about~~ 135 mM.

~~11. The pharmaceutical formulation of claims 1 to 10, wherein the Abeta antibody is a monoclonal antibody comprising a heavy chain and a light chain.~~

~~12. The pharmaceutical formulation of claim 11, wherein the heavy chain of the Abeta antibody comprises a VH domain which comprises:~~

~~— a CDR1 comprising the amino acid sequence of Seq. Id. No. 4,~~

~~— a CDR2 comprising the amino acid sequence of Seq. Id. No. 5,~~

~~— a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 6.~~

~~13. The pharmaceutical formulation of claim 11 or 12, wherein the light chain of the Abeta antibody comprises a VL domain which comprises:~~

~~— a CDR1 comprising the amino acid sequence of Seq. Id. No. 7,~~

~~— a CDR2 comprising the amino acid sequence of Seq. Id. No. 8,~~

~~— a CDR3 sequence comprising the amino acid sequence of Seq. Id. No. 9.~~

~~14~~12. The pharmaceutical formulation of claims ~~12~~ to ~~13~~11, wherein the VH domain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 2 and the VL domain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 3.

~~15~~13. The pharmaceutical formulation of claims ~~11~~ to ~~14~~12, wherein the heavy chain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 10.

~~16~~14. The pharmaceutical formulation of claims ~~11~~ to ~~15~~13, wherein light chain of the Abeta antibody comprises the amino acid sequence of Seq. Id. No. 11.

~~17~~15. The pharmaceutical formulation of claims ~~11~~ to ~~16~~14, wherein the monoclonal Abeta antibody is a mixture of mono-glycosylated Abeta antibodies and double-glycosylated Abeta antibodies, wherein the mono-glycosylated antibody comprises a glycosylated asparagine (Asn) at position 52 of Seq. Id. No. 2 in the VH domain of one antibody binding site and wherein the double-glycosylated antibody comprises a glycosylated asparagine (Asn) at position 52 of Seq. Id. No. 2 in the VH domain of both antibody binding sites and whereby said mixture comprises less than 5% of an antibody being non-glycosylated at position 52 of Seq. Id. No. 2 in the VH domain.

~~18~~16. ~~Use of t~~The pharmaceutical formulation of claims 1 to ~~17~~15 for use in the subcutaneous administration of the Abeta antibody.

~~19. The invention as herein described.~~