

STABLE LIQUID FORMULATIONS

TECHNICAL FIELD

The application relates to the development of a stable liquid formulation or composition suitable for pharmaceutical use and to the use of said formulation or composition in therapy, and in particular for intra venous or subcutaneous delivery of a therapeutic antibody.

SEQUENCE LISTING

The present application is filed with a Sequence Listing in electronic form. The entire contents of the sequence listing are hereby incorporated by reference.

BACKGROUND

Several diseases are thought to be caused by the abnormal folding and aggregation of disease-specific proteins. These proteins can accumulate into pathologically diagnostic accumulations, known as amyloids, which are visualized by certain histologic stains. Amyloids are thought to elicit inflammatory responses and have multiple negative consequences for the involved tissues. In addition, smaller aggregates of abnormally folded protein may exist and exert cytotoxic effects.

Transthyretin (TTR) is one of the many proteins that are known to misfold and aggregate (*e.g.*, undergo amyloidogenesis). Transthyretin-related amyloidosis encompasses two forms of disease: familial disease arising from misfolding of a mutated or variant TTR, and a sporadic, non-genetic disease caused by misaggregation of wild-type TTR. The process of TTR amyloidogenesis can cause pathology in the nervous system and/or heart, as well as in other tissues.

Antibodies recognizing Transthyretin have been described in several patent applications including WO2016/120810, WO2016/120811, WO2016/120809 and WO2018/007923. Treatment of diseases and disorders related to Transthyretin misfolding and/or misassemble has further been explored in such as WO2022/094152. Development of a lyophilized formulation has been described in WO2019/108689 in which three formulations comprising a) 20 mM histidine, 240 mM sucrose and 0.04% poloxamer 188, having a pH of 6.0, b) 20 mM succinate, 240 mM sucrose and 0.02% polysorbate 20, having a pH of 6.0 and c) 20 mM histidine, 205 mM trehalose and 0.02% polysorbate 20, having a pH of 6.0 were characterized and found to have good performance. The testing in WO2019/108689 was focused on development of a lyophilized formulation, which is an acceptable solution, but

since the processes of lyophilization and reconstitution are cumbersome, a stable liquid formulation is more attractive to ease the treatment burden and convenience for the patient.

To be suitable as a pharmaceutical formulation the liquid composition should ensure good stability of the antibody included, i.e., satisfactory physical and chemical stability.

5 Liquid formulations with a high-concentration antibody are likely to suffer from an increased viscosity which renders injections difficult, and it is thus further important that the formulation has an acceptable viscosity which is normally considered to be below 15-30 mPa*s depending on bioprocessing and administration.

SUMMARY

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 The present application provides antibody formulations including combinations of pharmaceutically acceptable excipients balancing physical and chemical stability, viscosity, and tonicity as further described below. Minimal particle formation (visible and subvisible) is preferred together with low levels of high molecular weight proteins (HMWP) i.e. aggregated
15 antibodies. Another parameter of interest is chemical stability, as formation of various isoforms and degradation products formed during storage can result in an inhomogeneous composition and thus a formulation providing a high degree of stability is preferred.

 The present invention discloses liquid formulations of an anti-Transthyretin antibody that demonstrate superior properties in terms of high stability.

20 The inventors have found that combining a buffer, a sugar, L-Arginine, L-Methionine and a surfactant provides a stable liquid formulation and it is further preferred that the liquid formulation includes a buffer providing a pH in the range of from 5 to 7 or such as about 5.5 to about 6.5.

25 In one aspect the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody,
- b) a buffer,
- c) a sugar,
- d) L-Arginine,
- 30 e) L-Methionine and
- f) a surfactant, and

wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

 In one aspect the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody present at a concentration within the range from about 10 mg/mL to about 200 mg/mL,
b) a buffer present at a concentration within the range from about 10 mM to about 50 mM,
c) a sugar present at a concentration within the range from about 50 mM to about 250 mM,
5 d) L-Arginine at a concentration within the range from about 50 mM to about 300 mM,
e) L-Methionine at a concentration within the range from about 10 mM to about 20 mM, and
f) a surfactant present at a concentration within the range from about 0.2 mg/mL to 5.0 mg/mL, and
wherein the pH of the formulation is within the range from about 5 to about 7.

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In one aspect the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody present at a concentration within the range from about 10 mg/mL to about 200 mg/mL,
b) a buffer present at a concentration within the range from about 10 mM to about 50 mM,
15 c) a sugar present at a concentration within the range from about 50 mM to about 250 mM,
d) L-Arginine at a concentration within the range from about 50 mM to about 150 mM,
e) L-Methionine at a concentration within the range from about 10 mM to about 20 mM, and
f) a surfactant present at a concentration within the range from about 0.2 mg/mL to 1.0 mg/mL, and
20 wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

In one aspect the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody present at a concentration within the range from about 10 mg/mL to about 200 mg/mL;
25 b) a buffer, wherein the buffer is histidine,
c) a sugar, wherein the sugar is sucrose,
d) 50-150 mM L-Arginine,
e) 10-20 mM L-Methionine and
30 f) a surfactant, wherein the surfactant is Poloxamer 188 (PX188), Polysorbate 80 (PS80) or Polysorbate 20 (PS20),
and wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

In one aspect the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody present at a concentration within the range from about 10 mg/mL to about 200 mg/mL;
- b) a buffer at a concentration of about 10-50 mM, wherein the buffer is histidine;
- c) a sugar present at a concentration within the range from about 20 mM to about 200 mM, wherein the sugar is sucrose;
- 5 d) L-Arginine at a concentration within the range from about 50 mM to about 150 mM;
- e) L-Methionine at a concentration within the range from about 10 mM to about 20 mM; and
- f) a surfactant present at a concentration within the range from about 0.2 mg/mL to 1.0 mg/mL, wherein the surfactant is Poloxamer 188 (PX188), Polysorbate 80 (PS80) or
- 10 Polysorbate 20 (PS20),
- and wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

In one aspect the invention relates to a liquid formulation comprising:

- a) 10-200 mg/mL anti-Transthyretin antibody,
- 15 b) 10-50 mM histidine,
- c) 20-200 mM sucrose,
- d) 50-150 mM L-Arginine
- e) 10-50 mM L-Methionine, and
- f) 0.2-2.0 mg/mL PX188,
- 20 and wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

In one embodiment the formulation is essentially free of sodium chloride.

In some embodiments, the sugar is sucrose, and the surfactant is PX188.

- In some embodiments, the sugar is sucrose at a concentration of around 50 mM and
- 25 the surfactant is PX188 at a concentration of around 1.0 mg/mL (0.1 % w/v).

In some formulations, 100 mM arginine is present. In some formulations, L-Methionine is at a concentration of 3.0 mg/mL (20 mM). In some formulations, pH is 6.0.

In one embodiment, the anti-Transthyretin antibody comprises;

- i. a heavy chain variable region comprising three CDRs, wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is SEQ ID NO.: 4 and
- 30 ii. a light chain variable region comprising three CDRs, wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3 is SEQ ID NO.: 8

- In some further embodiments, the antibody comprises three heavy chain CDRs and
- 35 three light chain CDRs, wherein the three heavy chain CDRs are SEQ ID NOs 1, 2 and 4,

respectively and wherein the three-light chain CDRs are SEQ ID Nos 6, 7 and 8, respectively.

In another embodiment, the heavy chain variable region has an amino acid sequence comprising any one of SEQ ID NOs:9-11 and the light chain variable region has an amino acid sequence comprising any one of SEQ ID NOs: 12-14.

In a further embodiment, the heavy chain has an amino acid sequence comprising SEQ ID NO.: 10 and the light chain has an amino acid sequence comprising SEQ ID NO.: 14.

In one embodiment the heavy chain variable region is fused to a heavy chain constant region and the light chain variable region is fused to a light chain constant region. In one embodiment, the heavy chain constant region has the sequence of SEQ ID NO.: 17, 18 or 19 (wherein the C-terminal lysine is optional) and the light chain variable region is fused to a light chain constant region having the sequence of SEQ ID NO.:20 or 21.

In some embodiment, the antibody comprises a heavy chain of SEQ ID NO.: 15, and a light chain of SEQ ID NO.: 16.

The invention also provides a reconstituted formulation resulting from reconstituting a lyophilized formulation.

The invention also provides a method of treating or effecting prophylaxis of a subject having or at risk of a transthyretin-mediated amyloidosis, comprising administering to the subject an effective regime of a liquid pharmaceutical formulation as described herein.

DESCRIPTION

DEFINITIONS

The term "antibody" includes intact antibodies and binding fragments thereof. Typically, fragments compete with the intact antibody from which they were derived for specific binding to the target or antigen. Fragments include separate heavy chains, separate light chains, Fab, Fab', F(ab')₂, F(ab)_c, Fv, single chain antibodies, and single domain antibodies. The term "antibody" also includes a bispecific antibody.

The basic antibody structural unit is a tetramer of subunits. Each tetramer includes two identical pairs of polypeptide chains, each pair having one "light" chain (about 25 kDa) and one "heavy" chain (about 50-70 kDa). The amino-terminal portion of each chain includes a variable region of about 100 to 110 or more amino acids primarily responsible for antigen recognition. When initially expressed, this variable region is typically linked to a cleavable signal peptide. The variable region without the signal peptide is sometimes referred to as a

mature variable region. Thus, for example, a light chain mature variable region means a light chain variable region without the light chain signal peptide. The carboxy-terminal portion of each chain defines a constant region primarily responsible for effector function. A constant region can include any or all of a CH1 region, hinge region, CH2 region, and CH3 region.

5 Light chains are classified as either kappa or lambda.

 Heavy chains are classified as gamma, mu, alpha, delta, or epsilon, and define the antibody's isotype as IgG, including subtype IgG1, IgG2, IgG3 and IgG4, IgM, IgA, IgD and IgE, respectively. The heavy chains may further include one or more amino acid substitution compared to the wild type sequences.

10 Within light and heavy chains, the variable and constant regions are joined by a "J" region of about 12 or more amino acids, with the heavy chain also including a "D" region of about 10 or more amino acids.

 The (mature) variable regions of each light/heavy chain pair form the antibody binding site. Thus, an intact antibody has two binding sites. Except for bifunctional or
15 bispecific antibodies, the two binding sites are the same. The chains all exhibit the same general structure of relatively conserved framework regions (FR) joined by three hypervariable regions, also called complementarity determining regions or CDRs. The CDRs from the two chains of each pair are aligned by the framework regions, enabling binding to a specific epitope. From N-terminal to C-terminal, both light and heavy chains variable regions
20 comprise the regions FR1, CDR1, FR2, CDR2, FR3, CDR3 and FR4. The assignment of amino acids to each region is in accordance with the definitions of Kabat, *Sequences of Proteins of Immunological Interest* (National Institutes of Health, Bethesda, MD, 1987 and 1991), or Chothia & Lesk, *J. Mol. Biol.* 196:901-917 (1987); Chothia *et al.*, *Nature* 342:878-883 (1989) or CDRs can be defined by the alternative definitions in Table 1 below.

25 Kabat also provides a widely used numbering convention (Kabat numbering) in which corresponding residues between different heavy chains or between different light chains are assigned the same number.

Loop	Kabat	Chothia	Composite of Chothia & Kabat	AbM	Contact
L1	L24--L34	L24--L34	L24--L34	L24--L34	L30--L36
L2	L50--L56	L50--L56	L50--L56	L50--L56	L46--L55
L3	L89--L97	L89--L97	L89--L97	L89--L97	L89--L96
H1	H31--H35B	H26--H32..H34*	H26--H35B*	H26--H35B	H30--H35B
H2	H50--H65	H52--H56	H50--H65	H50--H58	H47--H58
H3	H95--H102	H95--H102	H95--H102	H95--H102	H93--H101

Table 1: Conventional Definitions of CDRs Using Kabat Numbering. *CDR-H1 by Chothia can end at H32, H33, or H34 (depending on the length of the loop). This is because the Kabat numbering scheme places insertions of extra residues at 35A and 35B, whereas Chothia numbering places them at 31A and 31B. If neither H35A nor H35B (Kabat numbering) is present, the Chothia CDR- H1 loop ends at H32. If only H35A is present, it ends at H33. If both H35A and H35B are present, it ends at H34.

Antibodies are frequently referred to as monoclonal antibodies, wherein the term monoclonal originally referred to the antibody preparation originating from “one clone”, to differentiate from polyclonal antibodies which includes a plurality of antibodies to the antigen.

As antibodies are now mostly produced by recombinant methods a liquid formulation comprising an antibody herein refers to a highly homogeneous population of antibody produced by a host cell engineered to express the antibody of interest. The formulation therefore substantially homogeneous i. e., the individual antibodies are identical except for possible naturally occurring variations that may be present in very minor amounts.

It is generally understood that the C-terminal lysine of a heavy chain constant region is frequently removed during manufacture, and thus a preparation of an antibody may be in-homogenous with regards to such C-terminal lysine.

The content of the formulations is described herein with references to various ranges of the ingredients. Unless the context dictates the contrary, all ranges set forth herein should be interpreted as being inclusive of their endpoints and open-ended ranges should be

interpreted to include only commercially practical values. Further, all ranges of values should be considered as inclusive of intermediate values unless the context indicates the contrary.

As used herein, the term “about” or “approximately”, when used together with a numeric value (e.g., 150 mM or 10 %), refers to a range of numeric values that can be slightly less and slightly more than the number given. In some embodiments, the term “about” herein means ± 10 % of the value referred to. In some embodiments, the term “about” herein means ± 5 % of the value referred to. In some embodiments, the term “about” herein means ± 2 % of the value referred to. In some embodiments, the term “about” herein means ± 1 % of the value referred to. For example, “about 5” refers to a range of numeric values that are 10 %, 5 %, 2 %, or 1 % less or more than 5, e.g., ranges of 4.5 to 5.5, or 4.75 to 5.25, or 4.9 to 5.1, or 4.95 to 5.05.

The term “room temperature” as used herein, means a temperature between 15 °C and 30°C, such as between 20 °C and 30°C, between 20 °C and 25 °C or such as at 20°C, 25°C or 30°C. As pH measurements viscosity measurements are dependent on the temperature, and the temperature for viscosity measurements are as stated and generally measured at 5°C and/or 20°C.

Antigen

Transthyretin (TTR) is a 127-amino acid, 55 kDa serum and cerebrospinal fluid transport protein primarily synthesized by the liver. It has also been referred to as prealbumin, thyroxine binding prealbumin, ATTR, and TBPA. In its native state, TTR exists as a tetramer. In homozygotes, the tetramers comprise identical 127-amino-acid beta-sheet-rich subunits. In heterozygotes, the TTR tetramers are made up of variant and/or wild-type subunits, typically combined in a statistical fashion.

The established function of TTR in the blood is to transport *holo*-retinol binding protein. Although TTR is the major carrier of thyroxine (T_4) in the blood of rodents, utilizing binding sites that are orthogonal to those used for *holo*-retinol binding protein, the T_4 binding sites are effectively unoccupied in humans.

TTR is one of at least thirty different human proteins whose extracellular misfolding and/or misassembly (amyloidogenesis) into a spectrum of aggregate structures is thought to cause degenerative diseases referred to as amyloid diseases. TTR undergoes conformational changes in order to become amyloidogenic. Partial unfolding exposes stretches of largely uncharged hydrophobic residues in an extended conformation that efficiently misassemble into largely unstructured spherical aggregates that ultimately undergo conformation conversion into cross-beta sheet amyloid structures.

Unless otherwise apparent from context, reference to transthyretin (TTR) or its fragments or domains includes the natural human amino acid sequences including isoforms, mutants, and allelic variants thereof. Exemplary TTR polypeptide sequences are designated by Accession Numbers P02766.1 (UniProt), AAB35639.1 (GenBank), AAB35640.1

5 (GenBank), and ABI63351.1 (GenBank). Residues are numbered according to Swiss Prot P02766.1, with the first amino acid of the mature protein (*i.e.*, not including the 20 amino acid signal sequence) designated residue 1. In any other TTR protein, residues are numbered according to the corresponding residues in P02766.1 on maximum alignment.

10 Transthyretin (TTR) amyloidosis is a systemic disorder characterized by pathogenic, misfolded TTR and the extracellular deposition of amyloid fibrils composed of TTR. TTR amyloidosis is generally caused by destabilization of the native TTR tetramer form (due to environmental or genetic conditions), leading to dissociation, misfolding, and aggregation of TTR into amyloid fibrils that accumulate in various organs and tissues, causing progressive
15 dysfunction. See, *e.g.*, Almeida and Saraiva, *FEBS Letters* 586:2891-2896 (2012); Ando *et al.*, *Orphanet Journal of Rare Diseases* 8:31 (2013).

In humans, both wild-type TTR tetramers and mixed tetramers comprised of mutant and wild-type subunits can dissociate, misfold, and aggregate, with the process of amyloidogenesis leading to the degeneration of post-mitotic tissue. Thus, TTR amyloidoses
20 encompass diseases caused by pathogenic misfolded TTR resulting from mutations in TTR or resulting from non-mutated, misfolded TTR.

For example, senile systemic amyloidosis (SSA) and senile cardiac amyloidosis (SCA) are age-related types of amyloidosis that result from the deposition of wild-type TTR amyloid outside and within the cardiomyocytes of the heart. TTR amyloidosis is also the most
25 common form of hereditary (familial) amyloidosis, which is caused by mutations that destabilize the TTR protein. The TTR amyloidoses associated with point mutations in the TTR gene include familial amyloid polyneuropathy (FAP), familial amyloid cardiomyopathy (FAC), and the rare central nervous system selective amyloidosis (CNSA). Patients with hereditary (familial) TTR amyloidosis are almost always heterozygotes, meaning that the
30 TTR tetramers are composed of mutant and/or wild-type TTR subunits, generally statistically distributed. Hereditary (familial) versions of TTR amyloidosis are generally autosomal dominant and are typically earlier onset than the sporadic diseases (SSA and SCA).

Liquid formulations

As described herein above the present invention concerns a liquid formulation comprising an anti-Transthyretin antibody and one or more pharmaceutical excipients, such as a buffer system, tonicity agent(s), chelating agent(s), stabilizer(s), surfactant(s) and
5 preservative(s). For convenience reference is made to Remington: *The Science and Practice of Pharmacy*, 20th edition, 2000.

In one embodiment the liquid formulation includes an antibody and one or more pharmaceutical acceptable excipient, such as a buffer, an isotonic agent, an amino acid, a stabilizer, a surfactant and optionally a preservative.

10 The group of **buffers** includes sodium acetate, sodium carbonate, citrate, glycylglycine, histidine, glycine, lysine, arginine, sodium dihydrogen phosphate, disodium hydrogen phosphate, sodium phosphate, and tris(hydroxymethyl)-aminomethan, bicine, tricine, malic acid, succinate, maleic acid, fumaric acid, tartaric acid, aspartic acid or mixtures thereof.

15 In one embodiment the liquid formulation comprises histidine as buffer.

The group of **isotonic agents** includes a salt (e.g. sodium chloride), a sugar such as mono-, di-, or polysaccharides, or water-soluble glucans, including for example fructose, glucose, mannose, lactose, sucrose, trehalose, dextran, or sugar alcohol, an amino acid, an alditol (e.g. glycerol (glycerine), 1,2-propanediol (propyleneglycol), 1,3-propanediol, 1,3-
20 butanediol) polyethyleneglycol (e.g. PEG400), or mixtures thereof. Sugar alcohol includes, for example, mannitol, sorbitol, inositol, galactitol, dulcitol, xylitol, and arabitol. The use of an isotonic agent in pharmaceutical compositions is well-known to the skilled person.

In one embodiment the liquid formulation comprises sucrose.

25 Liquid formulations may further comprise an **amino acid** as a further isotonic agent or to decrease aggregate formation of the antibody during storage of the composition. The formulation may include an amino acid or a combination of amino acids, where any given amino acid is present either in its free base form or in its salt form.

In one embodiment the amino acid(s) may be proteogenic amino acids, such as
30 arginine, glycine, lysine, isoleucine, aspartic acid, glutamic acid, tryptophan and threonine.

In one embodiment the amino acid(s) may be non-proteogenic amino acids, such as aminoguanidine, ornithine and N-monoethyl L-arginine, ethionine and buthionine and S-methyl-L cysteine.

In one embodiment the amino acid(s) may be a combination of proteogenic amino
35 acids and non-proteogenic amino acids.

Liquid formulation may further comprise **methionine (or other sulphuric amino acids or amino acid analogous)** to inhibit oxidation of methionine residues to methionine sulfoxide when the polypeptide acting as the therapeutic agent is a polypeptide comprising at least one methionine residue susceptible to such oxidation. Any stereoisomer of methionine (L or D) or combinations thereof can be used.

In a further embodiment of the invention the formulation further comprises a **surfactant**. Typical surfactants (with examples of trade names given in brackets []) are polyoxyethylene sorbitan fatty acid esters such as polyoxyethylene (20) sorbitan monolaurate [Tween 20], polyoxyethylene (20) sorbitan monopalmitate [Tween 40] or polyoxyethylene (20) sorbitan monooleate [Tween 80], poloxamers such as polyoxypropylene-polyoxyethylene block copolymer [Pluronic F68/poloxamer 188], polyethylene glycol octylphenyl ether [Triton X-100] or polyoxyethyleneglycol dodecyl ether [Brij 35].

In a further embodiment of the invention the formulation further comprises a pharmaceutically acceptable **preservative**. In a further embodiment of the invention the preservative is selected from the group consisting of phenol, m-cresol, methyl p-hydroxybenzoate, propyl p-hydroxybenzoate, 2-phenoxyethanol, butyl p-hydroxybenzoate, benzyl alcohol, chlorobutanol, benzoic acid, imidurea, chlorocresol, ethyl p-hydroxybenzoate, benzethonium chloride, or mixtures thereof. The use of a preservative in pharmaceutical compositions is well-known to the skilled person. For convenience reference is made to Remington: *The Science and Practice of Pharmacy*, 20th edition, 2000.

Antibody formulations

Several antibodies binding to transthyretin (TTR) and humanized forms hereof have been described in WO2016/120810, WO2016/120811, WO2016/120809 and WO2018/007923.

The present application provides liquid formulations comprising such antibodies including, veneered or humanized forms hereof. In a preferred embodiment the antibody binds to residue 89-97 of TTR (SEQ ID NO.: 22). The present application further provides highly concentrated antibody formulations. A high concentrated antibody formulation is beneficial as it minimizes injection volume for patient convenience and suitable for intravenous administration or subcutaneous injection. However, a high concentration of antibody is likely to negatively affect viscosity, physical stability and formation of High Molecular Weight Proteins (HMWP). The present application provides antibody formulations including

combinations of pharmaceutically acceptable excipients balancing HMWP formation, viscosity, physical stability and tonicity as further described below.

Formulations of the invention comprise an anti-transthyretin antibody including chimeric, veneered or humanized version hereof, a buffer, one or more sugars and a surfactant, and have a pH within the range from about 5.5 to about 6.5.

The antibody formulation of the present invention is a liquid formulation, and in particular an aqueous formulation.

The formulations can be in liquid form. Liquid formulations can refer to a formulation before lyophilization or after reconstitution of a lyophilized formulation.

In general, components of a formulation other than water occur in the same relative proportions by weight or moles in a lyophilized formulation as in a liquid formulation prior to lyophilization. Likewise, components of the formulation after reconstitution with water are in general in the same relative proportions as in the formulation pre-lyophilization or the lyophilized formulation but the absolute concentrations can change in proportion to the relative volumes of the formulation pre and post reconstitution. Other components (besides water in liquid formulations) may include buffer, stabilizers, tonicity agent, viscosity lowering agent, surfactant, preservative and/or antioxidant which may be included to obtain a liquid formulation suitable for administration. Some excipients or groups of excipients may have multiple functions and thus effect the role of other excipients.

Antibody concentration

In liquid formulations according to the invention, the antibody can be present at a concentration within a range from about 10 to about 250 mg/mL, such as 20-225, 20-220, 20-175 or 50-150 mg/mL. In one embodiment the concentration is 10-250, 20-225 or 20-200 mg/mL, such as 25-190 mg/mL, 25-180 mg/mL, 25-175 mg/mL, 40-200 mg/mL, 40-180 mg/mL, 40-175 mg/mL or such as 40-170 mg/mL.

In one embodiment the antibody concentration is 10-200 mg/mL.

In some liquid formulations, the antibody concentration is 40-180 mg/mL, such as 40-60 mg/mL, 80-120 mg/mL or 130-180 mg/mL.

In some embodiments the liquid formulation comprises 25-75 mg/mL, 75-125 mg/mL or 125-175 mg/mL antibody.

In some embodiments the antibody is present at about 50 mg/mL.

In some embodiments the antibody is present at about 100 mg/mL.

In some embodiments the antibody is present at about 150 mg/mL.

Buffer and pH

Formulations include a buffer, such as, for example, citrate, histidine, phosphate or succinate, to confer specific pH range, such as from about 5.5 to about 7.0.

5 In a further embodiment the pH of the liquid formation is within the range of 5.5 to about 7.0, for example, a pH of 5.5, 5.6, 5.7, 5.8, 5.9, 6.0, 6.1, 6.2, 6.3, 6.4, 6.5, 6.6, 6.7, 6.8, 6.9, or 7.0. Some formulations have a pH of between about 5.5 to about 7.0, such as about 5.5 to about 6.5, or about 5.7 to about 6.3, or about 5.75 to about 6.25, or about 5.8 to about 6.2. Some formulations have a pH of about 6.0, such as a pH of 5.5-6.5, 5.6-6.4, 5.7-6.3 or 5.8-6.2. In one embodiment the formulation has a pH of about 6.5.

10 In some embodiments the buffer is citrate, histidine, phosphate or succinate.

In one embodiment the buffer is histidine. In such embodiment, histidine is present at a concentration within a range of about 10-30 mM, or 15-25 mM, for example at a concentration of about 10 mM, 20 mM or 25 mM. In one embodiment the buffer is L-histidine.

15 Further excipients

In further embodiments the formulation includes one or more additional excipients, such as a stabilizer, a tonicity agent, a viscosity lowering agent and/or a surfactant.

In one embodiment the formulation comprises one or more stabilizer(s). In one embodiment the formulation comprises one or more stabilizer(s), such as a sugar and or an amino acid.

20 In one embodiment the stabilizers may be a **sugar**.

In further embodiments the formulations include a sugar, such as, for example, trehalose or sucrose.

25 In further embodiments the sugar concentration is about 20-200 mM, 30-175 mM, or about 40-150 mM. In further embodiments the sugar concentration is about 20-100 mM, or about 30-70 mM, or about 40-60 mM, or about 50 mM.

In some embodiments the sugar is sucrose.

In further embodiments the sucrose concentration is about 20-200 mM, such as 30-175 mM, or about 40-150 mM.

30 In further embodiments the sucrose concentration is about 20-100 mM, such as about 30-70 mM, 40-60 mM, or about 50 mM.

In one embodiment an amino acid may be included in the formulation as a further stabilizer. In one embodiment the formulation includes **an amino acid**, such as L-Arginine, such as L-Arginine, HCl.

In some embodiment the concentration of L-Arginine is about 50-300 mM L-Arginine. In some embodiment the concentration of L-Arginine is about 50-150 mM L-Arginine, such as 60-140 mM, 70-140 mM, 70-130 mM, 70-120 mM, 80-120 mM or 90-110 mM L-Arginine. In one embodiment the formulation includes about 100 mM L-Arginine

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In one embodiment an amino acid may be included in the formulation as an **antioxidant**. In one embodiment the formulation includes an amino acid, such as L-Methionine.

In some embodiment the concentration of L-Methionine is about 5-50 mM L-Methionine, such as 10-44 mM, 15-45 mM, 15-40 mM, 15-35 mM, 15-30 mM or 15-25 mM L-Methionine. In one embodiment the formulation includes about 20 mM L-Methionine.

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In further embodiments the formulations include a **surfactant**, such as, for example, polysorbate 20 (PS20), polysorbate 80 (PS80) or a poloxamer, for example, poloxamer 188 (also known as PX188, PLURONIC F68 or FLOCOR). The surfactant can be present at a concentration within the range from about 0.01-0.2 %, 0.02-0.175 %, or 0.03%-0.15% (w/v). For example, the concentration can be 0.03%, 0.04%, 0.05%, 0.01%, 0.06%, 0.07%, 0.08%, 0.09%, 0.1%, 0.11%, 0.12%, 0.13%, 0.14% or 0.15 % (w/v).

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In one embodiment the concentration of the surfactant is from about 0.2 mg/mL to 5.0 mg/mL, such as from about 0.2 mg/mL to about 4.0, 3.0, 2.0 or 1.0 mg/mL.

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Poloxamers are nonionic triblock copolymers composed of a central hydrophobic chain of polyoxypropylene (poly(propylene oxide)) flanked by two hydrophilic chains of polyoxyethylene (poly(ethylene oxide)).

In one embodiment the surfactant is PS80 or PX188. In one embodiment the formulation includes 0.01%-0.15%, 0.02%-0.15%, or 0.03%-0.15% (w/v). In one embodiment the surfactant is PS80 and some formulations include about 0.04%, about 0.05% or about 0.1% w/v PS80.

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In one embodiment the surfactant is PX188 and some formulation include about 0.02%, about 0.04% or about 0.1% (w/v) PX188.

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An aspect of the invention relates to a liquid formulation comprising:

- a) an anti-Transthyretin antibody,
- b) a buffer,
- c) a sugar,
- d) L-Arginine,

35

e) L-Methionine and

f) a surfactant,

wherein the pH of the formulation is within the range from about 5.5 to about 6.5.

In some formulations, the buffer comprises histidine, such as, for example, a
5 histidine buffer.

In a further embodiment the liquid formulation includes

an anti-Transthyretin antibody as described herein above wherein the antibody is present
at a concentration within the range from about 20 mg/mL to about 200 mg/mL;

a) a buffer present at a concentration within the range from about 10 mM to about 50 mM,
10 wherein the buffer is histidine;

b) a sugar present at a concentration within the range from about 20 mM to about 200 mM,
wherein the sugar is sucrose;

c) L-Arginine at a concentration within the range from about 50 mM to about 150 mM;

d) L-Methionine at a concentration within the range from about 5 mM to about 50 mM; and

15 e) a surfactant present at a concentration within the range from about 0.2 mg/mL to 2.0
mg/mL, wherein the surfactant is Poloxamer 188 (PX188) or Polysorbate 80 (PS80) or
Polysorbate 20 (PS20);

wherein the formulation is characterized by a pH within the range from about 5.5 to about
6.5.

20 In a further embodiment the liquid formulation comprises

a) 20-200 mg/mL anti-Transthyretin antibody,

b) 10-50 mM histidine,

c) 20-200 mM sucrose,

d) 50-150 mM L-Arginine,

25 e) 5-30 mM L-Methionine and

f) 0.2-2.0 mg/mL surfactant,

wherein the formulation has a pH of 5.5-6.5.

In some such formulations, the sugar is sucrose, and the surfactant is PS20, PS80 or

PX188, for example PS80 at a concentration of 0.01% w/v or PX188 at a concentration of
30 0.1% w/v (1 mg/mL).

In some embodiments no sodium chloride is added during manufacturing and thus
only minimal amounts of sodium chloride receding from impurities of other ingredients may
remain and thus the formulation is essentially free of sodium chloride. In some embodiments
the concentration of sodium chloride is below 10 mM, such as below 5 mM, or such as below
35 2 mM.

Likewise, in a further embodiment the formulation is essentially free of mannitol or sorbitol or both mannitol and sorbitol.

Pharmaceutical compositions

5 Formulations intended for administration to humans are preferably made under good manufacturing practices (GMP) approved or approvable by the FDA or a regulatory agency for a country other than the United States, for example, the European Medicines Agency, for preparation of drugs for administration to humans.

10 The formulations are sterile, for example, as accomplished by sterile filtration using a 0.2 µm or a 0.22 µm filter.

 In one embodiment the liquid formulation is a pharmaceutical formulation.

 In one embodiment the formulation is sterile.

 Some such formulations are characterized by an osmolality in the range of 200-500 mOsm/kg, such as 250-400 mOsm/kg or 270-350 mOsm/kg.

15 In further embodiment the osmolality is in the range of about 270 mOsm/kg to about 350 mOsm/kg, such as, for example, about 335 mOsm/kg.

 Liquid formulations can be substantially isotonic implying an osmolality of about 260-350 mOsm/kg water. Some formulations have an osmolality of 290-330 mOsm/kg.

20 Anti-transthyretin antibody

 Several antibodies binding to transthyretin (TTR) and humanized forms hereof have been described in WO2016/120810, WO2016/120811, WO2016/120809 and WO2018/007923. Formulations of the invention comprise an anti-transthyretin antibody including chimeric, veneered or humanized version hereof as illustrated by the Examples
25 herein which test formulations of a humanized version of 14G8/9D5 disclosed in WO2016/120810. These antibodies bind epitopes comprising amino acid residues 89-97 (SEQ ID NO.: 22) of TTR buried in the native TTR tetramer exposed in monomeric misfolded aggregated or febril forms of TTS

 The binding specificity is primarily dependent on the complementarity determining
30 regions (CDRs).

 In one embodiment the formulation according to the invention includes an antibody comprising

 a heavy chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is SEQ ID NO.: 4,

a light chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3 is SEQ ID NO.: 8

In a further embodiment the formulation includes an antibody comprising a heavy chain variable region comprising three CDRs, wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 and CDR3 is SEQ ID NO.: 4
a light chain variable region comprising three CDRs, wherein CDR1 is SEQ ID NO.: 6, CDR2 is SEQ ID NO.: 7 and CDR3 is SEQ ID NO.: 8.

In a further embodiment the heavy chain variable region has an amino acid sequence comprising any one of SEQ ID NOs: 9-11.

In a further embodiment the light chain variable region has an amino acid sequence comprising any one of SEQ ID NOs: 12-14.

In another embodiment the antibody comprises a heavy chain variable region having an amino acid sequence comprising any one of SEQ ID NOs: 9-11 and a light chain variable region having an amino acid sequence comprising any one of SEQ ID NOs: 12-14.

In a further embodiment the formulation includes an antibody comprising a heavy chain variable region comprising the amino acid sequence of SEQ ID NO.: 10 and a light chain variable region comprising the amino acid sequence of SEQ ID NO.: 14.

In one embodiment the heavy chain variable region is fused to a heavy chain constant region and the light chain variable region is fused to a light chain constant region.

In one embodiment, the heavy chain constant region has the sequence of SEQ ID NO.: 17, 18 or 19 (wherein the C-terminal lysine is optional) and the light chain variable region is fused to a light chain constant region having the sequence of SEQ ID NO.: 20 or 21.

In a further embodiment the formulation includes an antibody comprising a heavy chain comprising the amino acid sequence set forth as SEQ ID NO.: 15 and a light chain comprising an amino acid sequence set forth as SEQ ID NO.: 16

In one embodiment the invention relates to a formulation comprising:

- a) an anti-Transthyretin antibody having
 - i. a heavy chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is SEQ ID NO.: 4,
 - ii. a light chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3 is SEQ ID NO.: 8
- b) a buffer,

- c) a sugar,
- d) L-Arginine,
- e) L-Methionine and
- f) a surfactant,

5 wherein the formulation has a pH of 5.5-6.5.

In a further embodiment the formulation comprises:

- a) 20-200 mg/mL anti-Transthyretin antibody having
 - i. a heavy chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is
10 SEQ ID NO.: 4,
 - ii. a light chain variable region comprising three complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3 is
SEQ ID NO.: 8

- b) 10-50 mM histidine,
- 15 c) 20-200 mM sucrose,
- d) 50-150 mM L-Arginine,
- e) 5-30 mM L-Methionine and
- f) 0.2-2.0 mg/mL surfactant,

wherein the formulation has a pH of 5.5-6.5.

20

In one embodiment the liquid formulation comprises

- a) about 50-150 mg/mL of an antibody comprising three heavy chain CDRs and three light chain CDRs, wherein the three heavy chain CDRs are SEQ ID NOs 1, 2 and 4, respectively and wherein the three light chain CDRs are SEQ ID NOs 6, 7 and 8
25 respectively,
 - b) a histidine buffer at a concentration of about 20 mM;
 - c) sucrose at a concentration of about 50 mM;
 - d) poloxamer, such as PX188, at a concentration of about 0.1%;
 - e) about 100 mM L-Arginine;
 - 30 f) about 20 mM L-Methionine
- and has a pH of about 6.0.

In a further embodiment, the formulation consist essentially of an antibody comprising three heavy chain CDRs and three light chain CDRs, wherein the three heavy
35 chain CDRs are SEQ ID NOs 1, 2 and 4, respectively and wherein the three light chain

CDRs are SEQ ID Nos 6, 7 and 8, respectively, and the antibody is present at a concentration of about 50-150 mg/mL, and about 20 mM histidine, about 50 mM sucrose, 100 mM L-Arginine, 20 mM L-Methionine and about 0.1 % w/v PX188.

In a preferred embodiment the liquid formulation consists essentially of

- 5 a) an antibody comprising a mature heavy chain having the amino acid sequence set forth as SEQ ID NO.: 15 and a mature heavy chain having the amino acid sequence set forth as SEQ ID NO.: 16, and
- b) about 20 mM histidine,
- c) about 50 mM sucrose,
- 10 d) 100 mM L-Arginine,
- e) 20 mM L-Methionine and
- f) about 0.1 % w/v PX188,
- wherein the formulation has a pH of about 5.8-6.2.

Method for preparing a pharmaceutical composition

15

As a starting point for preparation of a liquid composition the antibody may be provided in a solution, usually an aqueous solution. This starting solution is preferably highly concentrated to allow dilution during preparation of the formulation. In one embodiment the starting solution of the antibody is more than 100 mg/mL, such as 100-300 mg/mL, or such as 150-200 mg/mL.

20

The usual method of preparation is to dissolve the excipients in a buffer solution which includes each excipient in a concentration equal to or higher than the concentration of the final composition, which is obtained after admixing the excipient solution with the antibody preparation. Depending on the concentration of the antibody preparation (starting solution) an appropriate volume of this is included to reach the desired concentration of the final pharmaceutical compositions. Depending on the volume and concentration of the antibody solution further water may be added as well.

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The formulation is prepared by mixing the antibody solution with the excipient solution and if needed water is added to reach the total volume. The pH may be adjusted at different steps, such as one or more of; the antibody solution, the buffer solution and the final formulation.

30

The skilled person will know how to modify the methods in various ways such as to adjust concentrations of the buffer solution if a different mixing relationship is required or if one or more of the components of the final composition is already present in the antibody

solution, wherefore the concentration of hereof in the buffer solution should be adjusted. Finally, a step of sterile filtration is usually added. The examples herein illustrate such methods.

Methods of treatment

5

Pharmaceutical compositions are generally administered by parenteral administration which for liquid composition may be performed by subcutaneous, intramuscular, intraperitoneal or intravenous injection by means of a syringe, optionally a pen-like syringe. Alternatively, parenteral administration can be performed by means of an infusion pump.

10

As described in the background section anti-TTR antibodies are being tested for treatment and prevention of diseases related to the abnormal folding and aggregation of Transthyretin (TTR), such as Transthyretin-related amyloidosis.

An aspect of the invention relates to the use of a liquid formulation comprising an anti-TTR antibody as described herein in a method of treatment. A further embodiment relates to the use in a method of treating and/or preventing transthyretin-mediated amyloidosis. In further embodiments, the method encompasses administering to a subject in need, an effective amount or an effective regime of the formulation. In one embodiment the subject is a human suffering from transthyretin-mediated amyloidosis.

15

In one embodiment Transthyretin-related amyloidosis is a familial disease arising from misfolding of a mutated or variant TTR or a sporadic, non-genetic disease caused by mis-aggregation of wild-type TTR.

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In one embodiment the treatment targets the process of TTR amyloidogenesis in one or more tissue, such as in the nervous system and/or heart.

25

EMBODIMENTS

1. A liquid formulation comprising:
 - a) an anti-Transthyretin antibody,
 - b) a buffer,
 - c) a sugar,
 - d) L-Arginine,
 - e) L-Methionine,

30

f) a surfactant and

wherein the pH of the formulation is within the range from 5.0 to 7.0 or about 5.5 to about 6.5.

- 5 2. The formulation according to embodiment 1, wherein the composition comprises a buffer selected from the group consisting of citrate, histidine, phosphate and succinate.
3. The formulation according to any of the preceding embodiments, wherein the buffer concentration is 10-50 mM.
- 10 4. The formulation according to any of the preceding embodiments, wherein the buffer is histidine.
5. The formulation according to any of the preceding embodiments, wherein the composition wherein the formulation has a pH of 5.5-6.5, 5.6-6.4, 5.7-6.3 or 5.8-6.2.
- 15 6. The formulation according to any of the preceding embodiments, wherein the composition comprises a sugar selected from the group consisting of: mono-, di-, or polysaccharides, water-soluble glucans, such as fructose, glucose, mannose, lactose, sucrose, trehalose, dextran, and sugar alcohols.
- 20 7. The formulation according to any of the preceding embodiments, wherein the sugar is sucrose.
- 25 8. The formulation according to any of the preceding embodiments, wherein the composition comprises 20-200 mM of sugar.
9. The formulation according to any of the preceding embodiments, wherein the composition comprises 20-200 mM sucrose.
- 30 10. The formulation according to any of the preceding embodiments, wherein the composition comprises L-Arginine.
11. The formulation according to any of the preceding embodiments, wherein the composition comprises 50-300 mM L-Arginine.
- 35

12. The formulation according to any of the preceding embodiments, wherein the composition comprises L-Methionine.
- 5 13. The formulation according to any of the preceding embodiments, wherein the composition comprises 5-50 mM L-Methionine.
14. The formulation according to any of the preceding embodiments, wherein the composition comprises a surfactant selected from the group consisting of. Polysorbate 80
10 (PS80), Polysorbate 20 (PS20) and Poloxamer 188 (PX188).
15. The formulation according to any of the preceding embodiments comprising:
- a) 20-250 mg/mL anti-Transthyretin antibody,
 - b) 10-50 mM histidine,
 - 15 c) 20-200 mM sucrose,
 - d) 50-300 mM L-Arginine,
 - e) 5-50 mM L-Methionine,
 - f) 0.2-5.0 mg/mL surfactant.
- 20 16. The formulation according to any of the preceding embodiments comprising:
- g) 20-250 mg/mL anti-Transthyretin antibody,
 - h) 10-50 mM histidine,
 - i) 20-200 mM sucrose,
 - j) 50-150 mM L-Arginine,
 - 25 k) 5-50 mM L-Methionine,
 - l) 0.2-5.0 mg/mL surfactant.
17. The formulation according to any of the preceding embodiments, wherein the surfactant is Poloxamer 188 (PX188), Polysorbate 80 (PS80) or Polysorbate 20 (PS20).
30
18. The formulation according to any of the preceding embodiments, wherein the formulation is essentially free of sodium chloride.
19. The formulation according to any of the preceding embodiments, wherein the
35 composition comprises 10-40 mM, such as 10-30 mM, such as 15-25 mM histidine.

20. The formulation according to any of the preceding embodiments, wherein the formulation comprises 40-100 mM sucrose, such as 40-80 mM sucrose or 40-60 mM sucrose.
- 5 21. The formulation according to any of the preceding embodiments, wherein the formulation comprises 75-130 mM L-Arginine, such as 80-120 mM L-Arginine, such as 85-110 mM or 90-110 mM L-Arginine.
- 10 22. The formulation according to any of the preceding embodiments, wherein the formulation comprises 10-50 mM L-Methionine, such as 10-45 mM, 15-45 mM, 15-40 mM, 15-35 mM, 15-30 mM or 15-25 mM L-Arginine.
- 15 23. The formulation according to any of the preceding embodiments, wherein the surfactant is PX188 and wherein the concentration of PX188 is 0.2-2.0 mg/mL, such as 0.2-1.5 mg/mL, 0.3-1.4 mg/mL or 0.4-1.2 mg/mL.
24. The formulation according to any of the preceding embodiments, wherein the surfactant is PX188 and wherein the concentration of PX188 is about 0.2, 0.4 or about 1.0 mg/mL.
- 20 25. The formulation according to any of the preceding embodiments, wherein the surfactant is PX188 and wherein the concentration of PX188 is 0.7-1.5 mg/mL, such as 0.8-1.2 mg/mL or 0.9-1.1 mg/mL.
- 25 26. The formulation according to any of the preceding embodiments, comprising about 20 mM L-histidine, about 50 mM sucrose, about 100 mM L-Arginine, about 20 mM L-Methionine and about 1.0 mg/mL PX188.
27. The formulation according to any of the preceding embodiments, wherein the formulation has a pH of 5.5-6.5, 5.8-6.2 or about 6.0.
- 30 28. A liquid formulation comprising 10-200 mg/mL anti-Transthyretin antibody, further comprising:
- (a) 20 mM histidine and 0.4 mg/ml PX188 at pH 5.5;
- (b) 20 mM histidine and 0.4 mg/ml PX188 at pH 6.0;
- 35 (c) 20 mM histidine and 0.4 mg/ml PX188 at pH 6.5;

- (d) 20 mM histidine, 150 mM L-Arginine and 0.4 mg/ml PX188 at pH 5.5;
(e) 20 mM histidine, 150 mM L-Arginine and 0.4 mg/ml PX188 at pH 6.0;
(f) 20 mM histidine, 150 mM L-Arginine and 0.4 mg/ml PX188 at a pH of 6.5;
(g) 20 mM histidine, 50 mM L-Arginine, and 0.4 mg/mL PX188 at pH 6.0;
5 (h) 20 mM histidine, 150 mM L-Arginine and 0.2 mg/mL PX188 at pH 6.0;
(i) 20 mM histidine, 150 mM L-Arginine and 0.4 mg/ ml PX188 at pH 6.0;
(j) 20 mM histidine, 150 mM L-Arginine, and 1 mg/mL PX188 at pH 6.0;
(k) 20 mM histidine and 0.5 mg/mL PS80 at pH 6.0;
(l) 20 mM histidine, 150 mM L-Arginine, and 0.4 mg/mL PS80 at pH 6.0;
10 (m) 20 mM histidine, 150 mM L-Arginine, and 0.5 mg/mL PS80 at pH 6.0;
(n) 20 mM histidine, 150 mM L-Arginine, and 1 mg/mL PS80 at pH 6.0;
(o) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose and 0.4 mg/mL PX188 at pH 6.0;
(p) 20 mM histidine, 150 mM L-Arginine, 1.5 mg/mL methionine and 0.4 mg/mL PX188 at
15 pH 6.0;
(q) 20 mM histidine, 150 mM L-Arginine, 3.0 mg/mL methionine and 0.4 mg/mL PX188 at pH 6.0;
(r) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 1.5 mg/mL methionine and 0.4 mg/mL PX188 at pH 6.0;
20 (s) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 3.0 mg/mL methionine and 0.4 mg/mL PX188 at pH 6.0; or
(t) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 3.0 mg/mL methionine and 1 mg/mL PX188 at pH 6.0.
- 25 29. A liquid formulation comprising 10-200 mg/mL anti-Transthyretin antibody, further comprising:
(p) 20 mM histidine, 150 mM L-Arginine, 1.5 mg/mL methionine and 0.4 mg/mL PX188, at pH 6.0;
(q) 20 mM histidine, 150 mM L-Arginine, 3.0 mg/mL methionine and 0.4 mg/mL PX188 at
30 pH 6.0;
(r) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 1.5 mg/mL methionine, and 0.4 mg/mL PX188 at pH 6.0;
(s) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 3.0 mg/mL methionine and 0.4 mg/mL PX188 at pH 6.0; or

(t) 20 mM histidine, 100 mM L-Arginine, 50 mM sucrose, 3.0 mg/mL methionine and 1 mg/mL PX188 at pH 6.0.

5 30. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody binds an epitope comprising amino acid residues 89-97 (SEQ ID NO.: 22) of TTR.

31. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody comprises

10 i. a heavy chain variable region comprising complementarity determining regions (CDRs) CDR wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is SEQ ID NO.: 4 and

ii. a light chain variable region comprising complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3

15 is SEQ ID NO.: 8.

32. The formulation according to any of the preceding claims, wherein the formulation comprises

a) about 50-150 mg/mL of an antibody comprising three heavy chain CDRs and three light

20 chain CDRs, wherein the three heavy chain CDRs are SEQ ID NOs 1, 2 and 4, respectively and wherein the three light chain CDRs are SEQ ID NOs 6, 7 and 8 respectively,

b) a histidine buffer at a concentration of about 20 mM;

c) sucrose at a concentration of about 50 mM;

25 d) poloxamer, such as PX188, at a concentration of about 0.1%;

e) about 100 mM L-Arginine;

f) about 20 mM L-Methionine

and has a pH of about 6.0.

30 33. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody is a humanized, chimeric or veneered antibody.

34. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody is an intact antibody or an antigen binding fragment.

35

35. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody comprises a heavy chain variable region comprising amino acid sequence of SEQ ID NO.: 10, and a light chain variable region comprising amino acid sequence of SEQ ID NO.: 14.
- 5
36. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody comprises a heavy chain constant region comprising amino acid sequence of SEQ ID NO.: 17, 18 or 19 and a light chain constant region comprising amino acid sequence of SEQ ID NO.: 20 or 21.
- 10
37. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody comprises a heavy chain comprising amino acid sequence of SEQ ID NO.: 15 and a light chain comprising amino acid sequence of SEQ ID NO.: 16.
- 15
38. The formulation according to any of the preceding embodiments, wherein the anti-Transthyretin antibody is present at a concentration of 20-250 mg/mL, such as 25-175 mg/mL, such as about 50, 100, or 150 mg/mL.
39. The formulation according to any of the preceding embodiments, wherein the formulation
- 20 is an aqueous liquid formulation.
40. The formulation according to any of the preceding embodiments, wherein the formulation is a pharmaceutical composition.
41. The formulation according to any of the preceding embodiments for use in a method of treatment.
- 25
42. The formulation according to any of the preceding embodiments for use in a method of treating and/or preventing transthyretin-mediated amyloidosis.
- 30
43. A method of treating and/or preventing transthyretin-mediated amyloidosis, comprising administering to a subject in need, an effective amount of the formulation according to any of the preceding embodiments.

44. A method of treating and/or preventing transthyretin-mediated amyloidosis, comprising administering to a subject in need, an effective regime of the formulation according to any of the preceding embodiments.

Examples

Analytical procedures

High Molecular Weight Proteins (HMWP) by SE-HPLC:

5

The amount of HMWP in the formulations was analyzed by SE-HPLC using isocratic elution on a size-exclusion column and subsequent UVdetection. The HMWP peak area relative to the total area was calculated and expressed in percent.

10 Acidic iso-forms:

The amount of acidic iso-forms was measured by imaged capillary isoelectric focusing (iCIEF). The method can separate proteins based on their isoelectric point. The percentage of total acidic isoforms is reported relative to the total peak area.

15

Sub-visible particles:

The amount of sub-visible particles was determined by Micro-Flow Imaging (MFI). Amount of sub-visible particles in the size > 5 µm, > 10 µm and > 25 µm was measured.

20

Oxidized forms:

The amount of oxidised forms were determined by RP-UHPLC. Oxidised forms is the sum of separate oxidised forms.

25

Viscosity:

Viscosity is measured with the Initium RheoSense instrument, where viscosity is calculated from a pressure drop in a microfluidic channel as the solution is moved under a specific flowrate. Measurements are carried out at 5 °C with sheer rates between 500-10.000 s⁻¹.

30

Purity:

The purity was determined by capillary electrophoresis – sodium dodecyl sulphate (CE-SDS) under non-reduced conditions. Non-reduced conditions monitor intact IgG and lower molecular weight assembly states of light and heavy chains. IgG purity was calculated relative to the total peak area.

Example 1: Formulation testing

Formulation testing was conducted with an anti-Transthyretin antibody having the mature heavy chain sequence of SEQ ID NO.: 15 (wherein C-terminal lysine is optional) and the mature light chain sequence of SEQ ID NO.: 16 produces recombinantly in CHO cells according to standard methods.

Sample preparation: The drug substance was formulated in histidine buffer. The drug substance contains a higher concentration of the active ingredient to allow for dilution to the target drug product concentration. The drug substance was thawed and mixed gently to ensure a homogenous solution. The thawed drug substance was diluted with a solution of excipients to generate the drug product. After mixing, the drug product solution was sterile filtrated, and 1 ml solution was filled in 2 ml vials.

22 formulations were tested including 100 mg/mL (formulations # 1-19) or 150 mg/mL (formulations # 20-22) antibody, L-histidine buffer in a pH range of from about 5.5 to about 6.5 with and without certain sugars, surfactants, antioxidants and other excipients. The compositions of 22 formulations are listed in Table 1.1.

#	pH	L-histidine (mM)	L-Arginine, HCl (mM)	NaCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Polysorbate 80 (mg/mL)	Other excipients
1	5.5	20	-	-	-	0.4	-	-
2	5.5	20	150	-	-	0.4	-	-
3	5.5	20	-	150	-	0.4	-	-
4	5.5	20	-	-	240	0.4	-	-
5	6.0	20	-	-	-	0.4	-	-

#	pH	L-histidine (mM)	L-Arginine, HCl (mM)	NaCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Polysorbate 80 (mg/mL)	Other excipients
6	6.0	20	150	-	-	0.4	-	-
7	6.0	20	-	150	-	0.4	-	-
8	6.0	20	-	-	240	0.4	-	-
9	6.5	20	-	-	-	0.4	-	-
10	6.5	20	150	-	-	0.4	-	-
11	6.5	20	-	150	-	0.4	-	-
12	6.5	20	-	-	240	0.4	-	-
13	6.0	20	-	-	-	-	0.5	-
14	6.0	20	150	-	-	-	0.5	-
15	6.0	20	-	150	-	-	0.5	-
16	6.0	20	-	-	240	-	0.5	-
17	6.0	20	-	-	-	0.4	-	240 mM trehalose
18	6.0	20	-	-	-	0.4	-	270 mM sorbitol
19	6.0	20	-	-	-	0.4	-	3 mg/mL (20 mM) methionine
20	6.0	20	-	-	-	0.4	-	-
21	6.0	20	50	-	-	0.4	-	-
22	6.0	20	150	-	-	0.4	-	-

Table 1.1 Liquid formulations of an anti-TTR antibody. Formulations # 1-19 include 100 mg/mL and formulations # 20-22 includes 150 mg/mL antibody.

The particles formation was assessed by visual inspection and by Micro-Flow

- 5 Imaging (sub-visible particles) and the viscosity of the formulations was tested at 5 °C at time zero.

- 10 The formulations were stored for 14 days at 40 °C. The amount of High Molecular Weight Proteins (HMWP) in the formulations were analysed by SE-HPLC. The increase in HMWP (%) during the 14 days storage at 40°C and the viscosity of the formulations at time zero are provided in Table 1.2 below.

#	Antibody concentration (mg/mL)	pH	Poloxamer 188 (mg/mL)	Other excipients (mM)	Viscosity at 5 °C (mPa*s)	HMWP (%)
1	100	5.5	0.4	-	7.08	1.3
2	100	5.5	0.4	150 Arginine	7.34	-0.5
3	100	5.5	0.4	150 NaCl	7.54	3.2
4	100	5.5	0.4	240 Sucrose	8.22	-0.8
5	100	6.0	0.4	-	5.77	1.0
6	100	6.0	0.4	150 Arginine	5.06	0.8
7	100	6.0	0.4	150 NaCl	7.21	3.0
8	100	6.0	0.4	240 Sucrose	8.26	0.9
9	100	6.5	0.4	-	6.14	1.1
10	100	6.5	0.4	150 Arginine	6.66	0.7
11	100	6.5	0.4	150 NaCl	6.13	1.9
12	100	6.5	0.4	240 Sucrose	8.71	0.4
17	100	6.0	0.4	240 trehalose	9.90	1.1
18	100	6.0	0.4	270 sorbitol	8.77	1.1
19	100	6.0	0.4	20 methionine	5.74	0.8
20	150	6.0	0.4	-	18.1	1.7
21	150	6.0	0.4	50 Arginine	11.7	1.3

#	Antibody concentration (mg/mL)	pH	Poloxamer 188 (mg/mL)	Other excipients (mM)	Viscosity at 5 °C (mPa*s)	HMWP (%)
22	150	6.0	0.4	150 Arginine	10.8	1.1

Table 1.2. Viscosity at time zero and change in HMWP (%) after 14 days for formulations 1-12 and 17-22 (including 0.4 mg/mL poloxamer 188).

#	Antibody concentration (mg/mL)	pH	Polysorbate 80 (mg/mL)	Other excipients (mM)	Viscosity at 5 °C (mPa*s)	HMWP (%)
13	100	6.0	0.5	-	6.33	1.1
14	100	6.0	0.5	150 Arginine	7.20	0.5
15	100	6.0	0.5	150 NaCl	6.41	1.5
16	100	6.0	0.5	240 Sucrose	8.79	-0.4

Table 1.3. Viscosity at time zero and change in HMWP (%) after 14 days for formulations 13-16 (including 0.5 mg/mL polysorbate 80).

- 5
- 10
- 15
- 20
- The results showed that higher viscosity was observed for formulations with 150 mg/mL antibody (formulation 20-22) as compared with formulations with 100 mg/mL antibody.
- Addition of 240 mM sucrose, 240 mM trehalose or 270 mM sorbitol increased viscosity of the formulations. Furthermore, the viscosity of formulations with pH 5.5 was slightly higher than formulations with pH 6.0. Meanwhile, addition of 50 mM or 150 mM L-Arginine, HCl lowered viscosity in particular for formulations with 150 mg/mL antibody (# 21-22).
- With respect to HMWP formation, addition of 150 mM NaCl increased the amounts of HMWP, while addition of 150 mM L-Arginine lowered the amount of HMWP in formulations with 100 mg/mL antibody or 150 mg/mL antibody. Addition of methionine also decreased HMWP amount in formulation 19.
- Furthermore, it was observed that formulations containing NaCl (formulation 3, 7, 11 and 15) had high particle counts at time zero (data not shown) but in general no or limited

signs of particle formation during storage for 14 days at 40°C were observed (data not shown).

Example 2: Further formulation testing

- 5
- Based on the results from the initial formulation testing, additional formulations were evaluated. This additional formulation testing was performed with the same antibody as used in Example 1. 20 formulations containing 50 mg/mL or 150 mg/mL antibody, 20 mM L-histidine buffer at pH 6.0 with and without certain sugars, surfactants, antioxidants and other excipients were prepared. The compositions of 20 formulations are listed in Table 2.1.
- 10
- Sample preparation: Drug substance is formulated in 20 mM L-histidine buffer. The drug substance contained a higher concentration of the active ingredient to allow for dilution to the target drug product concentration. The drug substance was provided as a frozen solution. The drug substance was thawed and mixed gently to ensure a homogenous solution. The thawed drug substance was diluted with a solution of excipients to give the
- 15
- aimed formulation and mixed. For formulations with methionine, addition of methionine was performed in an individual a last step by adding a small volume of a concentrated methionine solution to the otherwise final formulation. After mixing the final formulation solution was sterile filtrated through 0.22 µm filter and 1 ml is filled in 2 ml vials.

#	Antibody concentration (mg/mL)	Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Poly- sorbate 80 (mg/mL)	Methio- nine (mg/mL)
1	50	100	50	0.4	-	-
2	50	150	-	0.4	-	-
3	150	100	50	0.4	-	-
4	150	150	-	0.4	-	-
5	150	150	-	-	-	-
6	150	150	-	0.2	-	-
7	150	150	-	1.0	-	-
8	50	150	-	-	-	-
9	50	150	-	0.2	-	-
10	50	150	-	1.0	-	-

#	Antibody concentration (mg/mL)	Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Poly-sorbate 80 (mg/mL)	Methionine (mg/mL)
11	150	150	-	-	0.4	-
12	150	150	-	-	1	-
13	50	100	50	0.4	-	1.5
14	50	150	-	0.4	-	1.5
15	150	100	50	0.4	-	1.5
16	150	150	-	0.4	-	1.5
17	50	100	50	0.4	-	3.0
18	50	150	-	0.4	-	3.0
19	150	100	50	0.4	-	3.0
20	150	150	-	0.4	-	3.0

Table 2.1 Liquid formulations of an anti-TTR antibody.

The samples were analysed at time zero and after storage for 10 days at 50°C with respect to HMWP, viscosity, osmolality, acidic isoforms and particle formation.

- 5 Results for increase in HMWP (%) and viscosity for formulations 1-12, and osmolality for formulations 1-4 are provided in Table 2.2.

#	Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Polysorbate 80 (mg/mL)	HMWP (%) Diff. (0 to 10 days)	Viscosity (mPa*s at 20°C)	Osmolality (mOsm/kg)
1	100	50	0.4	-	2.4	1.86	266
2	150	-	0.4	-	3.2	1.88	299
8	150	-	0	-	2.9	1.88	307
9	150	-	0.2	-	2.9	1.84	339
10	150	-	1.0	-	3.0	1.92	NA
3	100	50	0.4	-	6.3	9.06	NA
4	150	-	0.4	-	6.3	8.89	NA
5	150	-	0	-	5.9	8.70	NA

6	150	-	0.2	-	5.4	8.68	NA
7	150	-	1.0	0	6.7	8.76	NA
11	150	-	-	0.4	6.1	8.63	NA
12	150	-	-	1.0	6.1	8.78	NA

Table 2.2. Change in HMWP (%) after 10 days, viscosity at time zero and osmolarity for a selection of the formulations of table 2.1 comprising either 50 mg/mL antibody (formulations # 1, 2, 8, 9 and 10) or 150 mg/mL antibody (formulations # 3-7, 11 and 12)

- 5 Results for HMWP (%) after 10 days storage at and amount of acidic isoforms after 10 days storage at 50 °C are provided in Table 2.3 for formulations with 0, 1.5 mg/mL or 3.0 mg/mL methionine.

#	Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Methionine (mg/mL)	HMWP (after 10 days)	Acidic isoforms (%) after 10 days
1	100	50	0.4	-	4.0	62.3
13	100	50	0.4	1.5	3.8	62.3
17	100	50	0.4	3.0	3.2	61.4
2	150	-	0.4	-	4.8	62.2
14	150	-	0.4	1.5	3.9	61.7
18	150	-	0.4	3.0	3.8	60.4
3	100	50	0.4	-	7.4	62.2
15	100	50	0.4	1.5	6.6	61.9
19	100	50	0.4	3.0	6.4	61.5
4	150	-	0.4	-	8.4	63.3
16	150	-	0.4	1.5	7.6	61.4
20	150	-	0.4	3.0	7.2	64.2

10 Table 2.3. HMWP (%) after 10 days and acidic isoforms for a selection of the formulations of table 3 comprising either 50 mg/mL antibody (formulations # 1, 13, 17, 2, 14, 18) or 150 mg/mL antibody (formulations # 3, 15, 19, 4, 16, 20)

The results have shown that highest increase of *HMWP* was observed for 150 mg/mL antibody compared to 50 mg/mL antibody and that the addition of 50 mM sucrose or methionine (1.5 mg/mL and 3.0 mg/mL) decreased the formation of HMWP. The results

further showed that addition of 50 mM sucrose and methionine (1.5 mg/mL and 3.0 mg/mL) had additive effect of lowering the formation of HMWP.

With respect to viscosity, increased concentration of antibody increased viscosity. All formulations of 150 mg/mL antibody exhibited viscosity lower than 10 mPa*s at 20 °C.

5 Furthermore, there were no or only minor viscosity differences comparing formulations containing 150 mM L-Arginine, HCl with formulations with 50 mM sucrose and 100 mM L-Arginine, HCl.

With respect to osmolality, formulations 1, 2, 8 and 9 were close to isotonic.

Regarding acidic isoforms, antibody concentration or presence of sucrose has no apparent
10 effect on the formation of acidic isoforms. In contrast, presence of methionine has a tendency to decrease the formation of acidic isoforms as measured with iCIEF suggesting reduced chemical degradation.

Example 3 – Stress tests

15 Formulations 1 to 12 from Example 2, were exposed to 2 and 5 freeze/thaw cycles. After the freeze/thaw process, the formulations were analysed with respect to the formation of HMWP and sub-visible particles (MFI).

Results for increase in HMWP (%) after the freeze/thaw cycles are provided in Table
3.1. The amount of sub-visible particles was based on Micro-Flow Imaging (MFI) and visual
20 inspections (photo not shown).

Formulation #	Antibody concentration (mg/mL)	L-Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Polysorbate 80 (mg/mL)	HMWP (%) Diff. (0 to 2 cycles)	HMWP (%) Diff. (0 to 5 cycles)
1	50	100	50	0.4	-	0	0
2	50	150	-	0.4	-	0	0
8	50	150	-	-	-	0	0
9	50	150	-	0.2	-	0	- 0.1
10	50	150	-	1.0	-	- 0.1	- 0.1
3	150	100	50	0.4	-	0	0

4	150	150	-	0.4	-	0	0
5	150	150	-	-	-	0	0
6	150	150	-	0.2	-	0	0
7	150	150	-	1.0	-	0	0
11	150	150	-	-	0.4	0	0
12	150	150	-	-	1.0	0	0

Table 3.1 Change in HMWP in response to stress testing by freeze/thaw cycles of formulation 1-12.

5 The results showed that after both 2 and 5 cycles of freeze/thaw, there was essentially no increase of HMWP for formulations 1-12.

 No increase in sub-visible particles was observed for formulations 1-4, 6-7 and 9-12. In contrast, formulations 5 and 8, which do not include surfactant, showed a substantial increase in particle formation (data not shown).

10 In a further stress test formulations 1-12 were agitated in vials stored in horizontal position with a frequency of 250 RPM for 1 week (7 days). At time zero and after 1 week the formulations were analysed with respect to the formation of HMWP and Sub-visible particles by Micro-Flow Imaging (MFI).

15 Results for increase in HMWP (%) as well as particle counts of particles larger than 5 µm, 10 µm and 25 µm are provided in Table 7. The number of particle-counts is an average of three determinations. The appearance of sub-visible particles was subject to visual inspection (photo not shown)

#	L-Arginine, HCl (mM)	Sucrose (mM)	Poloxamer 188 (mg/mL)	Polysorbate 80 (mg/mL)	HMWP (%) Diff. (0 to 7 days)	Particles > 5 µm	Particles > 10 µm	Particles > 25 µm
1	100	50	0.4	-	0.1	273	71	9
2	150	-	0.4	-	0	185	47	4
8	150	-	-	-	NA	NA	NA	NA
9	150	-	0.2	-	0.1	1400	597	152

10	150	-	1.0	-	0	360	70	8
3	100	50	0.4	-	0.6	638	194	24
4	150	-	0.4	-	0.5	824	319	41
5	150	-	-	-	NA	NA	NA	NA
6	150	-	0.2	-	0.5	1788	702	128
7	150	-	1.0	-	0.5	309	111	8
11	150	-	-	0.4	0.5	286	43	8
12	150	-	-	1.0	0.5	263	62	10

Table 3.2 Change in HMWP in response to stress testing by agitation for 7 days of formulation 1-12 comprising either 50 mg/mL antibody (formulations # 1, 2, 8, 9 and 10) or 150 mg/mL (formulations # 3-7, 11 and 12) antibody. NA. Formulations without surfactant where not analyzed, as particles were visible even before shaking and pronounced precipitation was observed shortly after initiation of shaking at 250 rpm; furthermore, high counts of sub-visible particles were present.

The results showed that the highest increase of HMWP was observed for formulations comprising 150 mg/mL antibody as compared to formulations comprising 50 mg/mL antibody. The results also showed that the addition of increasing amount of surfactant reduces particle formation.

In contrast, with the addition of surfactant, no precipitation was observed even after one week of shaking. The results have shown that the addition of Poloxamer 188 reduced the amount of sub-visible particles and that the 150 mg/mL antibody solutions exhibited the lowest counts of subvisible particles with 1 mg/mL Poloxamer 188 as compared to 0.2 and 0.4 mg/mL Poloxamer 188.

The additional experiments showed that a formulation having about 20 mM (3.1 mg/mL) L-Histidine, about 50 mM Sucrose (17.1 mg/mL), about 100 mM L-Arginine, HCl (21.1 mg/mL), about 20 mM L-Methionine (3.0 mg/mL) and about 0.1% w/v Poloxamer188 (1.0 mg/mL) gives a low formation rate of HMWP, low viscosity, low particle formation during physical stress and a suitable tonicity for 50-150 mg/mL antibody solutions.

Example 4 - Verification Studies

The liquid Formulation 1 and the liquid Formulation 2 were prepared as follows.

For Formulation 1, the drug product was manufactured by mixing, sterile filtration, filling in 20 ml vials, lyophilisation and capping of vials. After lyophilization, Formulation 1 was reconstituted with 4.9 ml Water For Injections (WFI) to arrive at a liquid composition comprising about 50 mg/mL antibody, about 3.1 mg/mL L-histidine (20 mM, about 82.2 mg/mL sucrose (240 mM), about 0.4 mg/mL PX188 (0.04 % (w/v) and a pH of 6.0.

For Formulation 2, drug substance was formulated in 20 mM histidine buffer. The drug substance contained a higher concentration of the active ingredient to allow for dilution to the target drug product concentration. The drug substance was provided as a frozen solution. The drug substance was thawed and mixed gently to ensure a homogenous solution. The thawed drug substance was diluted with a solution of excipients to give the aimed formulation. After mixing the final drug product solution was sterile filtrated and 20 ml was filled in 20 ml vials. Formulation 2 comprises about 50, 100 or 150 mg/mL antibody, 3.1 mg/mL L-histidine (20 mM), 17.1 mg/mL sucrose (50 mM), 21.1 mg/mL L-Arginine (100 mM), 3.0 mg/mL methionine (20 mM), 1.0 mg/mL Poloxamer 188 (0.1 % (w/v) and a pH of 6.0. The compositions of Formulation 1 and 2 are provided in Table 4.1.

	Formulation 1	Formulation 2
	mg/mL	mg/mL
Antibody concentration (mg/mL)	50	50, 100 and 150
L-Histidine (mM)	20	20
Sucrose (mM)	240	50
L-Arginine, HCl (mM)	NA	100
Methionine (mM)	NA	20
Poloxamer 188 (mg/mL)	0.4	1.0
pH	6.0	6.0

Table 4.1. Liquid formulations 1 and 2.

A stability test was performed to evaluate the stability of the liquid formulation during storage at 30 °C and samples collected at time zero and after 1 month and 3 months. Results for increase of HMWP, formation of oxidized forms, formation of acidic isoforms, decrease in purity and impact on potency after storage at 30°C for 1 month and 3 months as compared time zero are provided in Table 4.2.

	Formulation 1		Formulation 2					
Antibody conc. (mg/mL)	50		50		100		150	
Time point	1M	3M	1M	3M	1M	3M	1M	3M
HMWP (%)	0.4	0.6	0	0	0.1	0.1	0.2	0.3
Oxidised forms (%)	0.9	1.9	0.1	0.3	0	0.2	0	-0.3
Acidic isoforms (%)	4.0	7.9	1.5	6.9	4.7	9.7	4.4	8.9
Purity (%)	-0.7	-2.0	-0.8	-2.4	-0.6	-2.1	-0.8	-2.3
Potency (U/mg)	0.02	-0.08	0.02	0	-0.03	0	-0.15	-0.06

4.2. Results obtained for HMWP, formation of oxidized forms, formation of acidic isoforms, decrease in purity and impact on potency.

The robustness of light of Formulation 1 and Formulation 2 were also investigated.

- 5 The equipment used (Suntest XLS+ (II)) light incubator) was installed with a Xenon lamp equipped with a filter system consisting of a coated quartz glass dish, a window glass dish and a ID65 glass dish. The Xenon lamp includes ultraviolet, visible and infrared light and thus a simulation of full spectrum sunlight is achieved. The combination of the lamp and filter system provided a spectral distribution identical to the D65/ID65 standard defined in ISO 10977. Thus, the light source complies with section 1.B/option 1 in the ICH guideline Q1B. For the Xenon lamp used, 220 lux hours correspond to 1 watt hours/square meter (1 Wh/m²). 1.2 mill lux hours corresponds to approximately 5455 Wh/m² and an UV radiation of minimum 200 Wh/m². Program no. 1 in the Suntest XLS+ light incubator results in the specified light intensity of minimum 1.2 mill lux hours after 28 hours. Results for increase of HMWP, oxidized forms, acidic isoforms and purity after exposure to light are provided in Table 4.3:

	Formulation 1	Formulation 2		
Antibody (mg/mL)	50	50	100	150
HMWP (%)	2.8	1.1	1.6	2.1
Oxidised forms (%)	18.6	4.1	3.9	2.8
Acidic isoforms (%)	20.8	4.6	3.7	2.1
Purity (%)	-4.2	-1.5	-1.4	-1.8

4.3. Stability results obtained for formulation 1 and 2.

The results showed that Formulation 2 has improved stability at all three concentrations, increased robustness by impact of light and minimally impact on potency at all three concentrations as compared to Formulation 1.

5

A liquid pharmaceutical formulation suitable for injection should have high physical and chemical stability and a low viscosity. From the studies described above it is apparent that it is more attractive to use Arginine, HCl as viscosity lowering agent compared to sodium chloride which had a negative influence on physical stability (influence on particle formation, increased content of HMWP). The data further show that both sucrose and methionine, alone or in combination lowers the amount of HMWP. Finally, 1.0 mg/mL Poloxamer 188 reduces the amount of sub-visible particles, particular for liquid formulations with a high antibody concentration.

10

15

Example 5 – Impact of presence of chloride ions

To evaluate the impact of presence of chloride ions four different formulations were prepared, see table 5.1. Formulation #1 and 2 were prepared using Arginine, HCl. Formulation # 3 and 4 were prepared after buffer exchange of the drug substance to eliminate presence of chloride ions. Arginine was added and the amount of sucrose were doubled. pH adjustment was performed with glutamic acid. After formulation the drug product solutions were sterile filtrated, and 1 ml solution was filled in 2 ml vials.

20

Table 5.1 Overview of formulations

#	Anti- Transthyretin antibody (mg/ml)	L- Histidine (mM)	Methionine (mM)	Sucrose (mM)	Arginine (mM)	Poloxamer (mg/ml)	Glutamate (mM)
1	100	20	20	50	100 ^A	1.0	-
2	150	20	20	50	100 ^A	1.0	-
3	100	20	20	100	100 ^B	1.0	100
4	150	20	20	100	100 ^B	1.0	100

^AWith HCl - arginine concentration 100 mM

25

^BWithout HCl – arginine concentration 100 mM

Stability of the four formulations were followed for 3 months at 30°C. Results for Osmolality, increase in HMWP and level of sub-visible particles (MFI) are given in table 5.2.

Table 5.2 Results

#	Osmolality (mOsm/kg)	HMWP (%) Diff. (0 to 3 mths at 30°C	Particles > 5 µm		Particles > 10 µm		Particles > 25 µm	
			Time zero	3 mths at 30°C	Time zero	3 mths at 30°C	Time zero	3 mths at 30°C
1	309	0.1	233	917	44	451	4	76
2	333	0.2	182	634	54	256	10	42
3	394	0.1	27	669	8	172	0	4
4	425	0.2	31	703	8	237	4	31

5

Whereas addition of 150 mM NaCl was demonstrated to increase HMWP formation (Example 1) no further improvement was observed when chloride ions were completely removed by using glutamate for pH adjustment instead of HCl. No differences can be observed for any of the parameters investigated. The parameters Appearance, pH, purity, total fragments, isoforms and oxidized forms were also followed in stability confirming the above conclusion (data not shown).

10

While certain features of the invention have been illustrated and described herein, many modifications, substitutions, changes, and equivalents will now occur to those of ordinary skill in the art. It is, therefore, to be understood that the appended claims are intended to cover all such modifications and changes as fall within the true spirit of the invention.

15

CLAIMS

1. A liquid formulation comprising:
 - a) an anti-Transthyretin antibody,
 - 5 b) a buffer,
 - c) a sugar,
 - d) L-Arginine,
 - e) L-Methionine and
 - f) a surfactant, and
- 10 wherein the pH of the formulation is within the range from 5.0-7.0.
2. The formulation according to claim 1 comprising:
 - a) 20-250 mg/mL anti-Transthyretin antibody,
 - b) 10-50 mM histidine,
 - 15 c) 20-200 mM sucrose,
 - d) 50-300 mM L-Arginine,
 - e) 5-50 mM L-Methionine and
 - f) 0.2-5.0 mg/mL surfactant.
- 20 3. The formulation according to claim 1 or claim 2, wherein the surfactant is Poloxamer 188 (PX188), Polysorbate 80 (PS80) or Polysorbate 20 (PS20).
4. The formulation according to any of the preceding claims, wherein the formulation is essentially free of sodium chloride.
- 25 5. The formulation according to any of the preceding claims, wherein the formulation comprises 40-60 mM sucrose.
6. The formulation according to any of the preceding claims, wherein the surfactant is PX188 and wherein the concentration of PX188 is 0.2-2.0 mg/mL, such as 0.2-1.5 mg/mL, 0.3-1.4 mg/mL or 0.4-1.2 mg/mL such as about 0.2, 0.4 or 1.0 mg/mL.
- 30 7. The formulations according to any one of the preceding claims, wherein Methionine is present at a concentration of 10-50 mM L-Methionine, such as 10-45 mM, 15-45 mM, 15-

40 mM, 15-35 mM, 15-30 mM or 15-25 mM Methionine and wherein Arginine is present at a concentration of 50-150 mM, such as 75-125 mM L-Arginine.

8. The formulation according to any of the preceding claims, wherein the formulation
5 comprises about 20 mM L-histidine, about 50 mM sucrose, about 100 mM L-Arginine, about 20 mM L-Methionine and 1.0 mg/mL PX188.
9. The formulations according to any of the preceding claims, wherein the anti-Transthyretin antibody comprises
 - 10 i. a heavy chain variable region comprising complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 1, CDR2 is SEQ ID NO.: 2 or 3 and CDR3 is SEQ ID NO.: 4 and
 - ii. a light chain variable region comprising complementarity determining regions (CDRs) wherein CDR1 is SEQ ID NO.: 5 or 6, CDR2 is SEQ ID NO.: 7 and CDR3
15 is SEQ ID NO.: 8.
10. The formulation according to any of the preceding claims, wherein the anti-Transthyretin antibody comprises a heavy chain variable region comprising amino acid sequence of
20 SEQ ID NO.: 10, and a light chain variable region comprising amino acid sequence of SEQ ID NO.: 14.
11. The formulation according to any of the preceding claims, wherein the anti-Transthyretin antibody comprises a heavy chain comprising amino acid sequence of SEQ ID NO.: 15
25 and a light chain comprising amino acid sequence of SEQ ID NO.: 16.
12. The formulation according to any of the preceding claims, wherein the anti-Transthyretin antibody is present at a concentration of 40-180 mg/mL, such as about 50, 100, or 150 mg/mL.
- 30 13. The formulation according to any of the preceding claims, wherein the pH is about 5.5-6.5, 5.8-6.2 or about 6.0.
14. The formulation according to any of the preceding claims, wherein the formulation comprises

- a) about 50-150 mg/mL of an antibody comprising three heavy chain CDRs and three light chain CDRs, wherein the three heavy chain CDRs are SEQ ID NOs 1, 2 and 4, respectively and wherein the three light chain CDRs are SEQ ID NOs 6, 7 and 8 respectively,
- 5 b) a histidine buffer at a concentration of about 20 mM;
- c) sucrose at a concentration of about 50 mM;
- d) poloxamer, such as PX188, at a concentration of about 0.1%;
- e) about 100 mM L-Arginine;
- f) about 20 mM L-Methionine
- 10 and has a pH of about 6.0.
15. The formulation according to any of the preceding claims, where in the formulation is an aqueous pharmaceutical formulation.