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(54) Title: STABILIZED ANTIBODY SOLUTIONS

(57) Abstract: There is provided, *inter alia*, an aqueous solution comprising an antibody protein and a stabilizing mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol.

STABILIZED ANTIBODY SOLUTIONS

BACKGROUND OF THE INVENTION

When formulated as aqueous solutions, antibody proteins are susceptible to structural degradation during storage. The processes involved in protein degradation can be divided into physical (e.g. loss of quaternary, tertiary or secondary structure, aggregation, particle formation) and chemical (i.e. processes involving a covalent change such as deamidation, aspartate isomerization, oxidation, hydrolytic clipping etc.). Each of the degradants (e.g. soluble aggregated species, insoluble aggregated species and chemically modified variants) can impact the biological activity, toxicity or immunogenicity of the antibody protein.

Therefore, the level of all degradants has to be kept within the tight specifications that are set for each antibody protein product. The rates of the degradation processes depend on temperature and antibody proteins are generally more stable at lower temperatures. Consequently, commercial antibody products must typically be stored refrigerated. However, with increasing trend toward subcutaneous products that can be self-administered by the patient, there is a strong need to develop antibody protein products that can be used outside the cold chain, at least for a period of time, such as 2 weeks, such as 4 weeks, such as 12 weeks or more. The ability to store the product outside the cold chain often results in considerable improvement in convenience for the patient during the in-use period. Allowed excursions outside the cold chain can also significantly improve shipment logistics.

The present invention addresses the problem of instability of antibody proteins, in particular the problem of antibody protein degradation.

WO2006/0096488A2 (Pharmacia & Upjohn Company LLC) describes compositions of human IgG antibodies comprising a chelating agent, said to exhibit improved chemical and/or physical stability.

WO2013/114122A2 (Arecor Limited) describes an aqueous solution comprising an antibody protein at a concentration of at least about 10 mg/mL and an oligomer of ethyleneimine, wherein the number of repeating units of ethyleneimine (n) in the oligomer is in the range of n = 2-12.

WO2010/062896A1 (Abbott Laboratories) describes compositions and methods for inhibiting fractionation of immunoglobulins comprising a lambda light chain based on the observation that iron, in the presence of histidine, results in increased fragmentation of a recombinant fully human IgG molecule containing a lambda light chain due to cleavage in the hinge region.

SUMMARY OF THE INVENTION

The present invention addresses the problem of instability of antibody proteins. In one embodiment, the invention relates to an aqueous solution comprising an antibody protein and a stabilizing mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol. In one embodiment, the invention provides a method of stabilizing an antibody protein in an aqueous solution to storage comprising the step of adding to the solution a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol.

DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to the discovery that an aqueous solution of antibody protein can be stabilized by a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

The term “aqueous solution”, as used herein, refers to a solution in water, preferably distilled water, deionized water, water for injection, sterile water for injection or bacteriostatic water for injection. The aqueous solutions of the invention include dissolved antibody protein, a chelating agent which is a multi-anion, and a C3 polyol, and optionally, one or more additives and/or excipients. The aqueous solutions can also include one or more components, such as additives or excipients, which are partially dissolved or undissolved. The presence of such

component or components will result in a multi-phase composition, such as a suspension or an emulsion. Preferably, the aqueous solution of the invention is a homogeneous solution, as determined by eye or by light-scattering.

The term "antibody protein", as used herein refers to an antibody, an antibody fragment, an antibody conjugated to an active moiety, a fusion protein comprising one or more antibody fragments, such as an immunoglobulin Fc domain, or a derivative of any of the aforementioned. Examples of derivatives include conjugated derivatives e.g. an antibody or antibody fragment conjugated to another moiety. Such moieties include chemically inert polymers such as PEG. Preferred antibodies include monoclonal antibodies and polyclonal antibodies, preferably monoclonal antibodies. The monoclonal antibodies can be, for example, mammalian (e.g. murine) or avian, chimeric, for example, human/mouse or human/primate chimeras, humanized antibodies or fully human antibodies. Suitable antibodies include an immunoglobulin, such as IgG, including IgG_1, IgG_2, IgG_3 or IgG_4, IgM, IgA, such as IgA_1 or IgA_2, IgD, IgE or IgY. Suitable antibodies also include single chain antibodies. Also included are antibody fragments including Fc, Fab, Fab_2, ScFv fragments and the like. Also embraced are single domain antibodies including Nanobodies.

In certain embodiments, the antibody is fused or conjugated to an active molecule, such as a toxin or a chelating agent capable of binding a radioactive metal ion, such as ^{99}Tc, ^{111}Ir, ^{131}I or ^{90}Y. In such embodiments, the antibody typically functions as a targeting agent, for example, directing the active molecule to cells which display a certain cell surface protein.

Specific antibodies which can be formulated as described herein include, but are not limited to, infliximab (chimeric antibody, anti-TNFα), basiliximab (chimeric antibody, anti-IL-2), abciximab (chimeric antibody, anti- GpIIb/IIIa), daclizumab (humanized antibody, anti-IL-2), gemtuzumab (humanized antibody, anti-CD33), alemtuzumab (humanized antibody, anti-CD52), edrecolomab (murine Ig2a, anti-EpCAM), rituximab (chimeric antibody, anti-CD20), palivizumab (humanized antibody, anti-respiratory syncytial virus), trastuzumab (humanized antibody, anti- HER2/neu(erbB2) receptor), bevacizumab (humanized antibody, anti-VEGF), cetuximab (chimeric antibody, anti-EGFR), eculizumab (humanized antibody, anti-

complement system protein C5), efalizumab (humanized antibody, anti-CD 1Ia), ibritumomab (murine antibody, anti-CD20), muromonab-CD3 (murine antibody, anti- T cell CD3 receptor), natalizumab (humanized antibody, anti-α4 integrin), nimotuzumab (humanized IgGl, anti-EGF receptor), omalizumab (humanized antibody, anti-IgE), panitumumab (human antibody, anti-EGFR), ranibizumab (humanized antibody, anti-VEGF), 1-131 tositumomab (humanized antibody, anti-CD20), ofatumumab (human antibody, anti-CD-20), certolizumab (humanized antibody, anti-TNF-α), golimumab (human antibody, anti-TNFα) and denosumab (human antibody, anti-RANK ligand). Preferred antibodies include trastuzumab, rituximab, bevacizumab, cetuximab and ipilimumab. In one embodiment, the antibody is bevacizumab. In one embodiment, the antibody is not an anti-TNF-α antibody.

Other chimeric antibodies which can be formulated as described herein include bavituximab (anti-phosphatidylserine), brentuximab (anti-CD30), siltuximab (anti-IL-6), clenoliximab (anti-CD4), galiximab (anti-CD80), gomiliximab (anti-CD23), keliximab (anti-CD4), lumiliximab (anti-CD23), priliximab (anti-CD4), teneliximab (anti-CD40), vapaliximab (anti-VAP1), ecromeximab (anti-GD3), and pagibaximab (anti-staphylococcal lipoteichoic acid).

Other humanized antibodies which can formulated as described herein include epratuzumab (anti-CD22), afutuzumab (anti-CD20), bivatuzumab mertansine (anti-CD44), cantuzumab mertansine (anti-mucin), citatuzumab bogatox (anti-TACSTD1), dacetuzumab (anti-CD40), elotuzumab (anti-CD319), etaracizumab (anti-$\alpha_v\beta_3$-integrin), farletuzumab (anti-FRα), inotuzumab ozogamicin (anti-CD22), labetuzumab (anti-carcinoembryonic antigen), lintuzumab (anti-CD33), milatuzumab (anti-CD74), nimotuzumab (anti-EGFR), oportuzumab monatox (anti-EpCAM), pertuzumab (anti-HER2), sibrotuzumab (anti-FAP), tacatuzumab tetraxetan (anti-alpha-fetoprotein), tigatuzumab (anti-TRAIL-2), tucotuzumab celmoleukin (anti-EpCAM), veltuzumab (anti-CD20), aselizumab (anti-CD62L), apolizumab (anti-HLA-DRB), benralizumab (anti-CD125), cedelizumab (anti-CD4), epratuzumab (anti-CD22), erlizumab (anti—CD18), fontolizumab (anti-interferon-γ), mepolizumab (anti-IL5), ocrelizumab (anti-CD20), pascolizumab (anti-IL4), pexelizumab (anti-complement component 5), PRO-140 (anti-CCR5), reslizumab (anti-IL5), rontalizumab (anti interferon-α), rovelizumab (anti-CD11, CD18), siplizumab (anti-CD2), talizumab (anti-IgE), teplizumab

(anti-CD3), tocilizumab (anti-IL6R), vedolizumab (anti-$\alpha_4\beta_7$-integrin), visilizumab (anti-CD3), ibalizumab (anti-CD4), tefibazumab (anti-clumping factor A), tadocizumab (anti-$\alpha_{11b}\beta_3$-integrin), bapineuzumab (anti-amyloid-β), solanezumab (anti-amyloid-β), tanezumab (anti-NGF), urtoxazumab (anti-E. coli Shiga-like toxin II B subunit), felvizumab (anti-respiratory syncytial virus), motavizumab (anti- respiratory syncytial virus glycoprotein F) and lebrikizumab (anti-IL13).

Additional human antibodies which can be formulated as described herein include atorolimumab (anti-Rh factor), fresolimumab (anti-TGFβ-1, -2, and -3), lerdelimumab (anti-TGFβ-2), metelimumab (anti-TGFβ-1), morolimumab (anti-Rh factor), ipilimumab (anti-CTLA-4), tremelimumab (anti-CTLA-4), bertilimumab (anti-CCL11), zanolimumab (anti-CD4), briakinumab (anti-IL12, -23), canakinumab (anti-IL1β), ustekinumab (anti-IL12, -23), adecatumumab (anti-EpCAM), belimumab (anti-B cell activating factor), cixutumumab anti-IGF-1 receptor), conatumumab (anti-TRAIL-R2), figitumumab (anti-IGF-1 receptor), iratumumab (anti-CD30), lexatumumab (anti-TRAIL-R2), lucatumumab (anti-CD40), mapatumumab (anti-TRAIL-R4), necitumumab (anti-EGFR), olaratumab (anti-PDGF-Rα), pritumumab (anti-vimentin), robatumumab (anti-IGF-1 receptor), votumumab (anti-tumor antigen CTAA16.88), zalutumumab (anti-EGFR), stamulumab (anti-myostatin), efungumab (anti-fungal HSP90), exbivirumab (anti-hepatitis B surface antigen), foravirumab (anti- rabies glycoprotein), libivirumab (anti-hepatitis B surface antigen), rafivirumab (anti- rabies glycoprotein), regavirumab (anti-cytomegalovirus glycoprotein B), sevirumab (anti-cytomegalovirus), tuvirumab (anti-hepatitis B virus), panobacumab (anti-pseudomonas aeruginosa serotype IATS 011), raxibacumab (anti-anthrax toxin), ramucirumab (anti-VEGF-R2), and gantenerumab (anti-amyloid-β).

Fusion proteins comprising a fragment of an immunoglobulin molecule can also be formulated according to the invention. Suitable fusion proteins include proteins comprising an active protein domain fused to one or more immunoglobulin fragments, such as Fc domains. Such fusion proteins include dimeric proteins having monomeric units comprising an active protein domain, such as a soluble receptor or a receptor extracellular ligand binding domain, which is fused to an immunoglobulin Fc domain. Two Fc domains can associate via disulfide

bonds to form the dimeric protein. Such fusion proteins include etanercept, abatacept and belatacept.

Conjugated derivatives comprising antibodies (or one or more antibody fragments) and a chemically inert polymer such as PEG can also be formulated according to the invention. Such derivatives include certolizumab pegol.

The antibody protein can be isolated from natural sources or be a recombinant protein.

In certain embodiments, the antibody protein is substantially pure, that is, the composition comprises a single antibody protein and no substantial amount of any additional protein. In preferred embodiments, the antibody protein comprises at least 99%, preferably at least 99.5% and more preferably at least about 99.9% of the total protein content of the composition. In preferred embodiments the antibody protein is sufficiently pure for use as in a pharmaceutical composition.

The antibody protein is preferably a therapeutic antibody protein. Such an antibody protein has a desirable therapeutic or prophylactic activity and is indicated for the treatment, inhibition or prevention of a disease or medical disorder.

In one embodiment, antibody protein is a monoclonal antibody such as trastuzumab, rituximab, bevacizumab, cetuximab or ipilimumab. In another embodiment, the antibody protein is a fusion protein comprising an active protein domain fused to one or more immunoglobulin Fc fragments such as etanercept, abatacept or belatacept. In a further embodiment, the antibody is a derivative of an antibody protein and is a conjugated derivative comprising one or more antibodies or antibody fragments and a chemically inert polymer, such as certolizumab pegol.

The antibody protein is suitably present at a concentration of about 1 mg/mL to about 300 mg/mL, such as about 10 mg/mL to about 300 mg/mL, about 1 mg/mL to about 200 mg/mL or about 10 mg/mL to about 200 mg/mL.

The aqueous solution of the present invention comprises a chelating agent which is a multi-anion, as a stabilizing agent. By multi-anion is meant a species which has at least two anionic centres per molecule, at the particular pH of the solution. By chelating agent is meant an agent capable of complexing with metal ions such as calcium, magnesium, iron and/or zinc ions. Suitably, the chelating agent is capable of complexing with zinc ions. Typically, the multi-anion will have at least two anionic centres per molecule wherein the pH of the solution is between about pH 4.0 and about pH 8.0. In one embodiment, the chelating agent which is a multi-anion is ethylenediaminetetraacetate (EDTA). EDTA anion is preferably introduced into the aqueous solution in the form of a salt of ethylenediaminetetraacetatic acid, such as disodium or tetrasodium salt. Alternatively, it can be introduced in the form of ethylenediaminetetraacetatic acid with subsequent adjustment of pH to the required level. Further examples of a chelating agent which is a multi-anion include other chelating ions with four ionic centres such as ethylene glycol-bis (β-aminoethyl ether)-*N,N,N',N'*-tetraacetate (EGTA) and 1,2-bis(*o*-aminophenoxy)ethane-*N,N,N',N'*-tetraacetate (BAPTA) as well as citrate, pyrophosphate and alginate. The chelating agent which is a multi-anion may be employed as a suitable salt form (e.g. as a sodium salt), or as an acid form which forms a multi-anion in solution. A mixture of chelating agents may be used. The chelating agent which is a multi-anion has a stabilizing effect and is typically present at a concentration of about 0.1 mM to about 50 mM, such as about 0.1 mM to about 20 mM, e.g. about 0.1 mM to about 10 mM. Suitably, the chelating agent is not citrate.

In one embodiment, the chelating agent which is a multi-anion does not comprise cationic centres. In one embodiment, the chelating agent which is a multi-anion contains only anionic centres.

In one embodiment, the chelating agent which is a multi-anion comprises four anionic centres per molecule. In one embodiment, the chelating agent which is a multi-anion is tetradentate.

In one embodiment, the chelating agent which is a multi-anion has a logK metal binding stability constant with respect to zinc ion binding of >5.5 at 25 °C e.g. has a logK with respect

to zinc binding of >6, >6.5, >7, >7.5, >8, >8.5, >9, >9.5, >10, > 10.5, >11, >11.5, >12, >12.5, >13, >13.5, >14 or >14.5 at 25 °C. In one embodiment, the chelating agent which is a multi-anion has a logK with respect to zinc ion binding of between >5.5 and 15 at 25 °C e.g. has a logK with respect to zinc binding of between >6 and 15, between >6.5 and 15, between >7 and 15, between >7.5 and 15, between >8 and 15, between >8.5 and 15, between >9 and 15, between >9.5 and 15, between >10 and 15, between > 10.5 and 15, between >11 and 15, between >11.5 and 15, between >12 and 15, between >12.5 and 15, between >13 and 15, between >13.5 and 15, between >14 and 15 or between >14.5 and 15 at 25 °C. Metal binding stability constants listed in the National Institute of Standards and Technology reference database 46 (Critically Selected Stability Constants of Metal Complexes) can be used. The database typically lists logK constants determined at 25 °C e.g. citrate (logK = 4.93), EDTA (logK = 14.6), EGTA (logK = 12.6), BAPTA (logK = 10.26), pyrophosphate (logK = 8.71) and alginate (logK = 6.91). Suitably, the chelating agent which is a multi-anion has a logK with respect to zinc ion binding of between >5.5 and 15 at 25 °C.

The aqueous solution of the invention also comprises a C3 polyol as a stabilizing agent which is suitably selected from 1,2-propanediol (also known as propane-1,2-diol or propylene glycol) and glycerol (also known as 1,2,3-propanetriol, glycerin or glycerine). In one embodiment, the C3 polyol is 1,2-propanediol. In another embodiment, the C3 polyol is glycerol. In a further embodiment, the C3 polyol is a mixture of 1,2-propanediol and glycerol. The C3 polyol is suitably present at a concentration of about 100 mM to about 500 mM, such as about 150 mM to about 400 mM, or about 150 mM to about 300 mM. If more than one C3 polyol is present in the aqueous solution, then the concentration refers to the total concentration of C3 polyols.

Typically, the pH of the aqueous solution of the present invention is between about pH 4.0 and about pH 8.0, such as between about pH 5.0 and about pH 7.0 or between about pH 5.0 and about pH 6.5.

In one embodiment the aqueous solution of the invention further comprises a buffer in order to stabilise the pH of the formulation, which can also be selected to enhance antibody protein

stability. Suitably the buffer is selected from the group consisting of histidine, succinate, maleate, acetate, phosphate and TRIS. In an embodiment, the buffer is phosphate buffer.

In one embodiment, a buffer is selected to have a pK_a close to the pH of the composition; for example, histidine is suitably employed as a buffer when the pH of the composition is in the range 5.0-7.0. As another example, phosphate is suitably employed as a buffer when the pH of the composition is in the range 6.1-8.1. Alternatively, in another embodiment, the solution of the invention is further stabilised as disclosed in WO2008/084237A2, which describes a formulation comprising a protein and one or more additives, characterised in that the system is substantially free of a conventional buffer, i.e. a compound with an ionisable group having a pK_a within 1 unit of the pH of the formulation at the intended temperature range of storage of the composition, such as 25 °C. In this embodiment, the pH of the formulation is set to a value at which the formulation has maximum measurable stability with respect to pH; the one or more additives (displaced buffers) are capable of exchanging protons with the insulin compound and have pK_a values at least 1 unit more or less than the pH of the formulation at the intended temperature range of storage of the formulation. The additives may have ionisable groups having pK_a between 1 to 5 pH units, preferably between 1 to 3 pH units, most preferably from 1.5 to 2.5 pH units, of the pH of the aqueous formulation at the intended temperature range of storage of the composition (e.g. 25 °C). Such additives may typically be employed at a concentration of 0.5-10 mM e.g. 2-5 mM.

Typically, the buffer is present at a concentration of about 0.5 mM to about 50 mM, such as about 1 mM to about 20 mM, e.g. about 2 mM to about 5 mM.

The aqueous solutions of the invention may optionally comprise a surfactant. In one embodiment, the surfactant is a non-ionic surfactant such as an alkyl glycoside e.g. dodecyl maltoside; a polysorbate surfactant such as polysorbate 80 or polysorbate 20; an alkyl ether of polyethylene glycol e.g. selected from polyethylene glycol (2) dodecyl ether, polyethylene glycol (2) oleyl ether and polyethylene glycol (2) hexadecyl ether; a block copolymer of polyethylene glycol and polypropylene glycol, such as poloxamer 188, poloxamer 407, poloxamer 171 or poloxamer 185; or an alkylphenyl ether of polyethylene glycol, such as 4-

(1,1,3,3-tetramethylbutyl)phenyl-polyethylene glycol. Suitably the non-ionic surfactant is present at a concentration of about 10 μg/mL to about 2000 μg/mL, such as about 50 μg/mL to about 1000 μg/mL, e.g. about 100 μg/mL to about 500 μg/mL.

The aqueous solution of the invention may cover a wide range of osmolarity, including hypotonic, isotonic and hypertonic aqueous solutions. Suitably, the aqueous solution of the invention is substantially isotonic. In one embodiment, the aqueous solution of the invention is isotonic. Suitably, the osmolarity of the aqueous solution is selected to minimize pain according to the route of administration e.g. upon injection. Preferred aqueous solutions have an osmolarity in the range of about 200 mOsm/L to about 500 mOsm/L. Preferably, the osmolarity is in the range of about 250 mOsm/L to about 350 mOsm/L. More preferably, the osmolarity is about 300 mOsm/L.

Tonicity of the aqueous solution may be adjusted with a tonicity modifier. Tonicity modifiers may be charged or uncharged.

Examples of charged tonicity modifiers include salts such as a combination of sodium, potassium, magnesium or calcium ions, with chloride, sulfate, carbonate, sulfite, nitrate, lactate, succinate, acetate or maleate ions (especially sodium chloride or sodium sulphate, particularly sodium chloride). Amino acids such as glycine, histidine or arginine may also be used for this purpose. In one embodiment, the charged tonicity modifier is selected from the group consisting of sodium chloride, sodium sulphate, sodium acetate, sodium lactate, glycine, histidine and arginine. Such a charged tonicity modifier is typically present at a concentration of about 25 mM to about 500 mM, such as about 50 mM to about 250 mM, e.g. about 150 mM.

Examples of uncharged tonicity modifiers include sugars, sugar alcohols and other polyols, such as sucrose, trehalose, mannitol, raffinose, lactose, dextrose, sorbitol or lactitol, or polyethylene glycols such as PEG300 or PEG400. In one embodiment, the uncharged tonicity modifier is sucrose, trehalose, mannitol, sorbitol, PEG300 or PEG400. The C3 polyol which is a required component of the aqueous solution of the invention may function as an

uncharged tonicity modifier. However, reference to an aqueous solution of the invention "further" comprising an uncharged tonicity modifier is intended to refer to an additional, further component to be added to the solution. Thus, the aqueous solution may further comprise an uncharged tonicity modifier which is other than a C3 polyol, and in particular is other than 1,2-propanediol and glycerol. Such an uncharged tonicity modifier is typically present at a concentration of about 50 mM to about 1000 mM, such as about 100 mM to about 500 mM, e.g. about 300 mM.

The aqueous solution of the invention can optionally include a preservative, suitably selected from phenol, m-cresol, chlorocresol, benzyl alcohol, propylparaben, methylparaben, benzalkonium chloride and benzethonium chloride. When present, the preservative is at a concentration of about 0.01 mM to about 100 mM. A preservative selected from phenol, m-cresol, chlorocresol, benzyl alcohol, propylparaben, methylparaben may, for example, be present at a concentration of about 10 mM to about 100 mM, such as about 20 mM to about 80 mM e.g. about 25 mM to about 50 mM. A preservative selected from benzalkonium chloride and benzethonium chloride may, for example, be present at a concentration of about 0.01 mM to about 1 mM such as about 0.05 mM to about 0.5 mM e.g. about 0.05 mM to about 0.2 mM.

The present inventors have discovered that the stability of an antibody protein in an aqueous solution is improved by the addition of a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol. The addition of a chelating agent which is a multi-anion such as EDTA has been observed to enhance the stability of an antibody protein in an aqueous solution. Surprisingly, the stabilizing effect of the EDTA is further increased by the addition of a C3 polyol. Without wishing to be bound by theory it is believed that the stabilising effect of the chelating agent which is a multi-anion is due to a combination of (i) charge interactions with positively charged patches at the surface of the protein and (ii) elimination of trace metals that may catalyse degradation processes. Without wishing to be bound by theory it is believed that the additional stabilising effect of a C3 polyol is due to optimal hydrophobic and hydrogen bond interactions at the protein surface of the small polyols leading to tighter conformation and modified interfacial tension between the protein molecules, in turn leading

to lower exposure of reaction sites as well as lower probability of irreversible aggregation events.

In one embodiment, the ratio (mM/mM) of chelating agent which is a multi-anion to C3 polyol is between about 1:5 and about 1:500 e.g. between about 1:20 and about 1:200.

In one embodiment, the ratio (wt/wt) of antibody protein to chelating agent which is a multi-anion is between about 1:1 and about 500:1 e.g. between about 10:1 and about 200:1. In another embodiment, the ratio (wt/wt) of antibody protein to chelating agent which is a multi-anion is between about 10:1 and about 1000:1 e.g. between about 50:1 and about 200:1.

In one embodiment, the ratio (wt/wt) of antibody protein to C3 polyol is between about 1:5 and about 200:1 e.g. between about 1:1 and about 50:1. In another embodiment, the ratio (wt/wt) of antibody protein to C3 polyol is between about 1:2 and about 200:1 e.g. between about 2:1 and about 50:1.

The addition of a mixture of chelator which is a multi-anion, and a C3 polyol to an aqueous solution of antibody protein is expected to enhance the stability of the antibody protein, e.g. as shown in Example 1. The mixture of a chelating agent which is a multi-anion and a C3 polyol is thus referred to as a stabilizing mixture.

The "stability" of an antibody protein or a "stabilizing mixture" typically refers to a reduction of antibody protein degradation during storage. In one embodiment, "stability"/"stabilizing" refers to physical stability e.g. loss of quaternary, tertiary or secondary structure, aggregation or particle formation. In another embodiment, "stability"/"stabilizing" refers to chemical stability e.g. processes involving a covalent change such as deamidation, aspartate isomerization, oxidation or hydrolytic clipping.

It is expected that the addition of a mixture of a chelating agent which is a multi-anion, and a C3 polyol, to an aqueous solution comprising an antibody protein can enhance the stability of the antibody protein and in particular reduce the rate of antibody protein aggregation,

compared with the same solution lacking the chelating agent which is a multi-anion, and a C3 polyol, following storage under the same conditions for the same length of time.

The present invention thus provides a method of stabilizing an antibody protein in an aqueous solution to storage comprising the step of adding to the solution a mixture of (i) a chelating agent which is a multi-anion; and (ii) C3 polyol. Also provided is the use of a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol, for stabilizing an antibody protein in an aqueous solution to storage. All embodiments described hereinabove with reference to the aqueous solution of the invention apply equally to the method and use of the invention.

The method of the invention refers to "the step of adding to the solution a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol". It should be understood that the chelating agent which is a multi-anion and the C3 polyol can be added to the solution at the same time, or sequentially, and in any order (i.e. "the step" may actually include multiple steps).

Also provided is a method for inhibiting formation of high molecular weight species of an antibody protein in aqueous solution during storage, comprising the step of adding to the solution a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

Also provided is a method for inhibiting formation of visible particles in an aqueous solution of an antibody protein during storage, comprising the step of adding to the solution a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

Also provided is a method for inhibiting formation of related species of an antibody protein in aqueous solution during storage, comprising the step of adding to the solution a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

Also provided is a method for inhibiting deamidation of an antibody protein in aqueous solution during storage, comprising the step of adding to the solution a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

Also provided is a method for inhibition formation of low molecular weight degradation products in an aqueous solution of an antibody protein during storage, comprising the step of adding to the solution a mixture of a chelating agent which is a multi-anion, and a C3 polyol.

Also provided is the use of a mixture of a chelating agent which is a multi-anion, and a C3 polyol for inhibiting the formation of high molecular weight species of an antibody protein in aqueous solution during storage.

Also provided is the use of a mixture of a chelating agent which is a multi-anion, and a C3 polyol for inhibiting the formation of visible particles in an aqueous solution of an antibody protein during storage.

Also provided is the use of a mixture of a chelating agent which is a multi-anion, and a C3 polyol for inhibiting formation of related species of an antibody protein in aqueous solution during storage.

Also provided is the use of a mixture of a chelating agent which is a multi-anion, and a C3 polyol for inhibiting deamidation of an antibody protein in aqueous solution during storage.

Also provided is the use of a mixture of a chelating agent which is a multi-anion, and a C3 polyol for inhibiting formation of low molecular weight degradation products in an aqueous solution of an antibody protein during storage.

The term “high molecular weight species” as used herein, refers to any component of the antibody protein content which has an apparent molecular weight at least about double the molecular weight of the parent active antibody protein. That is, high molecular weight species are multimeric aggregates of the parent antibody protein. The multimeric aggregates may comprise the parent antibody protein molecules with considerably altered conformation or they may be an assembly of the parent protein units in the native or near-native conformation. The determination of high molecular weight species can be done using methods known in the

art, including size exclusion chromatography, electrophoresis, analytical ultracentrifugation/sedimentation velocity, light scattering, dynamic light scattering, static light scattering and field flow fractionation.

The term "low molecular weight degradation products" as used herein, refers to any component of the antibody protein content which has an apparent molecular weight less than the molecular weight of the parent active antibody protein. That is, low molecular weight degradation products are fragments of the parent antibody protein. The determination of high molecular weight species can be done using methods known in the art, including size exclusion chromatography, electrophoresis, analytical ultracentrifugation/sedimentation velocity, light scattering, dynamic light scattering, static light scattering and field flow fractionation.

The term "related species" as used herein, refers to any component of the antibody protein content formed by a chemical modification of the parent antibody protein, such as deamidated species or oxidised species. Related species are suitably detected by cation-exchange chromatography, reversed-phase chromatography or capillary electrophoresis.

Suitably an aqueous solution of the invention is sufficiently stable such that it remains substantially free of visible particles after storage at 30 °C for at least one, two or three months. Visible particles are suitably detected using the 2.9.20. European Pharmacepoeia Monograph (Particulate Contamination: Visible Particles).

Suitably the aqueous solution of the invention is sufficiently stable such that the concentration of related species remains low upon extended storage.

In one embodiment, the aqueous solution of the invention retains at least 95%, e.g. at least 96%, e.g. at least 97%, e.g. at least 98%, e.g. at least 99% parent antibody protein (by weight of total antibody protein) after storage at 30°C for one, two or three months. The percentage of antibody protein (by weight of total antibody protein) may be determined by size-exclusion

chromatography, cation-exchange chromatography, reversed-phase chromatography or capillary electrophoresis.

In one embodiment, the presence of the mixture of chelating agent which is a multi-anion, and a C3 polyol limits the increase in high molecular weight antibody protein species to no more than 5% (by weight of total antibody protein) after storage at 40°C for one month, suitably to no more than 3% and more suitably to no more than 2%. In one embodiment, the presence of a mixture of a chelating agent which is a multi-anion, and a C3 polyol limits the increase in high molecular weight antibody protein species to no more than 5% (by weight of total antibody protein) after storage at 2-8°C for up to two years, suitably to no more than 3% and more suitably to no more than 2%. Quantitation of high molecular weight species is as percent by weight of the total antibody protein in the aqueous solution.

In one embodiment, the presence of the mixture of a chelating agent which is a multi-anion, and a C3 polyol limits the increase in high molecular weight antibody protein species by at least 10%, preferably by at least 25%, and more preferably by at least 50% compared with an aqueous solution lacking the chelating agent which is a multi-anion, and the C3 polyol but otherwise identical, following storage under the same conditions and length of time.

In one embodiment, the presence of the mixture of a chelating agent which is a multi-anion, and a C3 polyol maintains an aqueous solution of an antibody protein free of visible aggregates while formation of visible aggregates is observed in an aqueous solution lacking the mixture of the chelating agent which is a multi-anion, and a C3 polyol but otherwise identical, following storage under the same conditions and for the same length of time. Quantification of visible aggregates can be performed by turbidity or other types of light scattering measurement.

Suitably, the aqueous solution of the invention comprises no more than 5% (by weight of total protein) high molecular weight species after storage at 40°C for at least one, two or three months. In one embodiment, the amount of high molecular weight species increases by no more than 5% (by weight of total antibody protein), preferably no more than 3%, after storage

at 40°C for at least one, two or three months. Quantitation of high molecular weight species is as percent by weight of the total antibody protein in the aqueous solution.

Suitably, the aqueous solution of the invention should exhibit an increase in high molecular weight species during storage which is at least 10% lower, preferably at least 25% lower, more preferably at least 50% lower, than an aqueous solution lacking a mixture of a chelating agent which is a multi-anion, and a C3 polyol but otherwise identical, following storage under the same conditions and length of time.

In one embodiment, the aqueous solution of the invention is a pharmaceutical composition suitable for administration of a therapeutic antibody protein to a subject in need thereof. Such compositions can be used in a method for administering the therapeutic protein to the subject.

In another embodiment, the invention provides a method for administering a therapeutic antibody protein to a subject in need thereof. The method comprises the step of administering an aqueous solution comprising an antibody protein, a chelating agent which is a multi-anion, and a C3 polyol. Preferably the composition is administered by intravenous, subcutaneous or intramuscular injection, or infusion. More preferably the composition is administered by subcutaneous injection.

In another embodiment, the invention provides a packaged pharmaceutical composition suitable for administration to a subject in need thereof. The pharmaceutical composition comprises an aqueous solution comprising an antibody protein, a chelating agent which is a multi-anion, and a C3 polyol. The pharmaceutical composition is preferably packaged in a vial suitable for introduction of a needle for removal of the solution. In one embodiment, the pharmaceutical composition is packaged in a glass vial with a rubber stopper. The packaged pharmaceutical composition can be provided as a kit, further comprising instructions for use and, optionally, a syringe suitable for intramuscular or subcutaneous administration. Alternatively, the packaged pharmaceutical composition can be provided in the form of a pre-filled disposable syringe suitable for intramuscular or subcutaneous administration. A pre-

filled auto-injector device would also be suitable for intramuscular or subcutaneous administration.

The term "pharmaceutically acceptable", as used herein, refers to components of a pharmaceutical composition which are suitable for the intended use and mode of administration to the body of a human or an animal, such as a mammal, without undue adverse consequences, such as toxicity, irritation, and allergic response and with a reasonable risk/benefit ratio.

ABBREVIATIONS

EDTA	ethylenediaminetetraacetate
PEG	polyethylene glycol
HMWS	high molecular weight specie
SEC	size exclusion chromatography
CEX	cation-exchange chromatography

EXAMPLES

Materials

EDTA disodium salt (Mw 372 Da), 1,2-propanediol (Mw 76 Da), glycerol (Mw 92 Da), mannitol (Mw 182 Da), NaCl (Mw 58 Da), trehalose (Mw 342 Da) were obtained from Sigma Aldrich.

Methods of assessing stability of an antibody protein

(a) Visual assessment

Visible particles are suitably detected using the 2.9.20. European Pharmacepoeia Monograph (Particulate Contamination: Visible Particles). The apparatus required consists of a viewing station comprising:

- a matt black panel of appropriate size held in a vertical position

- a non-glare white panel of appropriate size held in a vertical position next to the black panel
- an adjustable lampholder fitted with a suitable, shaded, white-light source and with a suitable light diffuser (a viewing illuminator containing two 13 W fluorescent tubes, each 525 mm in length, is suitable). The intensity of illumination at the viewing point is maintained between 2000 lux and 3750 lux.

Any adherent labels are removed from the container and the outside washed and dried. The container is gently swirled or inverted, ensuring that air bubbles are not introduced, and observed for about 5 s in front of the white panel. The procedure is repeated in front of the black panel. The presence of any particles is recorded.

The visual scores are ranked as follows:

Visual score 1: Clear solution, virtually free of particles

Visual score 2: ~ 5 very small particles

Visual score 3: ~10-20 very small particles

Visual score 4: 20-50 particles, including larger particles

Visual score 5: >50 particles, including larger particles

Whilst the particles in samples with visual scores 4 and 5 are clearly detectable on casual visual assessment under normal light, samples with visual score 1-3 generally appear as clear solutions on the same assessment. Samples with visual scores 1-3 are considered to be "Pass"; samples with visual score 4-5 are considered to be "Fail".

(b) Size exclusion chromatography (SEC)

The amount of high molecular weight species is measured using a 300×7.8 mm S3000 (or equivalent) size-exclusion column with a guard column. The mobile phase is potassium phosphate pH 6.5, with a flow rate of 0.4 ml/min, injection volume of 1 µl and detected at 210 and 280 nm. The results are expressed as % high molecular species (HMWS), i.e. sum of all peak areas corresponding to aggregated protein over the sum of all protein-related peaks on the chromatogram. A small time-point to time-point variability can be observed in terms of absolute values of %HMWS, for example due to repeated size-exclusion column use. However, within a given time-point the samples are tested using the column in the same

condition, so the values generated within the time-point represent a very good indication of the relative stability of the protein in the aqueous solutions tested.

(b) Cation-exchange chromatography chromatography (CEX)

The amount of related species is measured using a Protein-Pak Hi Res SP column. Mobile phase A is 20 mM sodium phosphate (pH 6.5); mobile phase B is 20 mM sodium phosphate + 0.5 M NaCl (pH 6.0). The following gradient elution is used: 0 min – 100% A, 4 min – 80% A, 10 min – 55% A, 12 min – 0% A. Flow rate of 1.0 ml/min; injection volume is 3 µl, with UV detection at 214 nm. The results are expressed as % main peak (i.e. native protein), % acidic species and % basic species. % Related species = % acidic species + % basic species.

Example 1

The effect of EDTA and C3 polyols on the stability of abatacept (125 mg/ml) was investigated. The effect was tested in a background solution containing sodium phosphate (5 mM) and polysorbate 80 (0.5 mg/ml). All formulations tested were adjusted to pH 6.5. Additional excipients in the formulations tested are shown in Table 1.

Table 1: Additional components in formulations of abatacept tested. All formulations contained abatacept (125 mg/ml), sodium phosphate (5 mM) and polysorbate 80 (0.5 mg/ml) and were adjusted to pH 6.5.

	NaCl (mM)	Trehalose (mM)	1,2-propanediol (mM)	EDTA (mM)
Formulation 1	150	0	0	0
Formulation 2	0	300	0	0
Formulation 3	0	0	300	0
Formulation 4	150	0	0	1
Formulation 5	150	0	0	10
Formulation 6	150	0	0	50
Formulation 7	0	300	0	1
Formulation 8	0	300	0	10
Formulation 9	0	300	0	50

Formulation 10	0	0	300	1
Formulation 11	0	0	300	10
Formulation 12	0	0	300	50

Stability of Formulations 1-12 (Table 1) was tested at 25 °C and 40 °C by visual assessment and size-exclusion chromatography (SEC). Results are shown in Tables 2 and 3. It was shown that in the absence of EDTA the stability of abatacept was slightly better in the presence of an uncharged tonicity modifier (trehalose or 1,2-propanediol) than in the presence of a charged tonicity modifier (NaCl). Addition of EDTA appeared to improve the stability of abatacept, both with respect to visual assessment and with respect to the formation of HMWS. The degree of improvement was greater in compositions comprising the uncharged species. Surprisingly, the degree of improvement was greater in compositions comprising a C3 polyol (1,2-propanediol) than in compositions comprising a larger polyol (trehalose). This indicates synergistic effect between EDTA and 1,2-propanediol. Whilst the highest level of EDTA tested (50 mM) resulted in the best stability with respect to high molecular weight species it also appeared to result in slightly worse visual score. This could be due to the fact that more soluble aggregates (i.e. HMWS) are converted to insoluble aggregates in the presence of high concentration of EDTA.

Table 2: Visual scores of abatacept Formulations 1-12 following storage at 25 °C and 40 °C. Visual score 1: clear solution, virtually free of particles; visual score 2: ~ 5 very small particles; visual score 3: ~10-20 very small particles; visual score 4: 20-50 particles, including larger particles; visual score 5: >50 particles, including larger particles

	T = 0 weeks	T = 10 weeks (25°C)	T = 10 weeks (40°C)
Formulation 1	1	4	5
Formulation 2	1	3	4
Formulation 3	1	3	4
Formulation 4	1	2	4
Formulation 5	1	2	5
Formulation 6	1	3	5

Formulation 7	1	2	3
Formulation 8	1	2	3
Formulation 9	1	3	3
Formulation 10	1	1	2
Formulation 11	1	1	1
Formulation 12	1	1	2

Table 3: Stability of abatacept (125 mg/ml) in Formulations 1-12 assessed by SEC. Formation of HMWS was assessed following storage at 25°C and 40°C.

	HMWS (%)		
	T = 0 weeks	T = 10 weeks (25°C)	T = 10 weeks (40°C)
Formulation 1	0.61	3.58	14.73
Formulation 2	0.60	3.21	12.72
Formulation 3	0.55	3.49	13.15
Formulation 4	0.59	3.18	13.78
Formulation 5	0.60	3.18	13.26
Formulation 6	0.54	3.06	13.00
Formulation 7	0.62	2.79	10.26
Formulation 8	0.58	2.83	10.44
Formulation 9	0.61	2.46	9.99
Formulation 10	0.60	1.99	8.97
Formulation 11	0.60	1.98	8.29
Formulation 12	0.57	2.10	8.13

Example 2

The effect of EDTA and polyols on stability of bevacizumab (25 mg/ml) was investigated at 40 °C. The effect was tested in a background solution containing sodium phosphate (5 mM) and polysorbate 20 (0.4 mg/ml). All formulations tested were adjusted to pH 6.2. Additional

excipients in the formulations tested are shown in Table 4. The stability was also compared to the composition of the currently marketed bevacizumab product (Avastin®).

Table 4: Additional components in formulations of abatacept tested. All formulations contained bevacizumab (25 mg/ml) and polysorbate 20 (0.5 mg/ml) and were adjusted to pH 6.2.

	Sodium phosphate (mM)	NaCl (mM)	Trehalose (mM)	Mannitol (mM)	Glycerol (mM)	EDTA (mM)
Formulation 1 = Composition of Avastin®	50.4		158			
Formulation 2	5	130				
Formulation 3	5		300			
Formulation 4	5			300		
Formulation 5	5				300	
Formulation 6	5	130				10
Formulation 7	5	130				50
Formulation 8	5				300	10
Formulation 9	5				300	50

Stability of Formulations 1-9 (Table 4) was tested at 40 °C by visual assessment. Results are shown in Table 5. Formulation 1 (i.e. the composition of Avastin®) resulted in visual score 5 following 10 weeks storage at 40 °C. Similarly, visual score 5 was reached in compositions containing 5 mM sodium phosphate and either charged tonicity modifier (NaCl) or an uncharged tonicity modifier (mannitol, trehalose or glycerol). Better visual score was observed in the presence of EDTA (10 or 50 mM). However, the use of EDTA in the presence of NaCl still resulted in worse visual scores than the use of EDTA in the presence of a C3 polyol (glycerol), indicating a synergistic effect between EDTA and the C3 polyol.

Table 5: Visual scores of bevacizumab Formulations 1-9 following storage at 40 °C. Visual score 1: clear solution, virtually free of particles; visual score 2: ~ 5 very small particles; visual score 3: ~10-20 very small particles; visual score 4: 20-50 particles, including larger particles; visual score 5: >50 particles, including larger particles.

	T = 0 weeks	T = 10 weeks (40°C)
Formulation 1	1	5
Formulation 2	1	5
Formulation 3	1	5
Formulation 4	1	5
Formulation 5	1	5
Formulation 6	1	3
Formulation 7	1	3
Formulation 8	1	1
Formulation 9	1	2

Throughout the specification and the claims which follow, unless the context requires otherwise, the word 'comprise', and variations such as 'comprises' and 'comprising', will be understood to imply the inclusion of a stated integer, step, group of integers or group of steps but not to the exclusion of any other integer, step, group of integers or group of steps.

While this invention has been particularly shown and described with references to preferred embodiments thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the scope of the invention encompassed by the appended claims. It should also be understood that the embodiments described herein are not mutually exclusive and that features from the various embodiments may be combined in whole or in part in accordance with the invention.

All publications, patents, patent applications, internet sites, and accession numbers/database sequences (including both polynucleotide and polypeptide sequences) cited are herein incorporated by reference in their entirety for all purposes to the same extent as if each

individual publication, patent, patent application, internet site, or accession number/database sequence were specifically and individually indicated to be so incorporated by reference.

CLAIMS

1. An aqueous solution comprising an antibody protein and a stabilizing mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol.

2. A method of stabilizing an antibody protein in an aqueous solution to storage comprising the step of adding to the solution a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol.

3. Use of a mixture of (i) a chelating agent which is a multi-anion; and (ii) a C3 polyol, for stabilizing an antibody protein in an aqueous solution to storage.

4. The aqueous solution of claim 1, method of claim 2, or use of claim 3, wherein the chelating agent is a chelating ion with four ionic centres.

5. The aqueous solution, method or use of any one of claims 1 to 4, wherein the chelating agent which is a multi-anion is EDTA.

6. The aqueous solution, method or use of any one of claims 1 to 5, wherein the chelating agent which is a multi-anion is present at a concentration of about 0.1 mM to about 50 mM, such as about 0.1 mM to about 20 mM, e.g. about 0.1 mM to about 10 mM.

7. The aqueous solution, method or use of any one of claims 1 to 6, wherein the C3 polyol is 1,2-propanediol.

8. The aqueous solution, method or use of any one of claims 1 to 6, wherein the C3 polyol is glycerol.

9. The aqueous solution, method or use of any one of claims 1 to 6, wherein the C3 polyol is a mixture of 1,2-propanediol and glycerol.

10. The aqueous solution, method or use of any one of claims 1 to 9, wherein the C3 polyol is present at a concentration of about 100 mM to about 500 mM, such as about 150 mM to about 400 mM, or about 150 mM to about 300 mM.

11. The aqueous solution, method or use of any one of claims 1 to 10, wherein the antibody protein is a therapeutic antibody protein.

12. The aqueous solution, method or use of any one of claims 1 to 11, wherein the antibody protein is an antibody, an antibody fragment, an antibody conjugated to an active moiety, a fusion protein comprising one or more antibody fragments, or a derivative of any of the aforementioned.

13. The aqueous solution, method or use of claim 12, wherein the antibody protein is a monoclonal antibody.

14. The aqueous solution, method or use of claim 13, wherein the monoclonal antibody is a murine antibody, a chimeric antibody, a humanized antibody or a human antibody.

15. The aqueous solution, method or use of claim 13, wherein the monoclonal antibody is selected from trastuzumab, rituximab, bevacizumab, cetuximab and ipilimumab.

16. The aqueous solution, method or use of claim 15, wherein the monoclonal antibody is bevacizumab.

17. The aqueous solution, method or use of claim 12, wherein the antibody protein is a fusion protein comprising an active protein domain fused to one or more immunoglobulin Fc fragments.

18. The aqueous solution, method or use of claim 12, wherein the antibody protein is etanercept, abatacept or belatacept.

19. The aqueous solution, method or use of claim 12, wherein the derivative is a conjugated derivative comprising one or more antibodies or antibody fragments and a chemically inert polymer.

20. The aqueous solution, method or use of claim 19, wherein the conjugated derivative is a certolizumab pegol.

21. The aqueous solution, method or use of any one of claims 1 to 20, wherein the antibody protein is present at a concentration of about 1 mg/mL to about 300 mg/mL, such as about 10 mg/mL to about 300 mg/mL, about 1 mg/mL to about 200 mg/mL or about 10 mg/mL to about 200 mg/mL.

22. The aqueous solution, method or use of any one of claims 1 to 21, wherein the pH of the solution is between about pH 4.0 and about pH 8.0, such as between about pH 5.0 and about pH 7.0 or between about pH 5.0 and about pH 6.5.

23. The aqueous solution, method or use of any one of claims 1 to 22, further comprising a buffer.

24. The aqueous solution, method or use of claim 23, wherein the buffer is selected from the group consisting of histidine, succinate, maleate, acetate, phosphate and TRIS.

25. The aqueous solution, method or use of claim 23 or claim 24, wherein the buffer is present at a concentration of about 0.5 mM to about 50 mM, such as about 1 mM to about 20 mM, e.g. about 2 mM to about 5 mM.

26. The aqueous solution, method or use of any one of claims 1 to 25, further comprising a non-ionic surfactant.

27. The aqueous solution, method or use of claim 26, wherein the non-ionic surfactant is an alkyl glycoside, such as dodecyl maltoside.

28. The aqueous solution, method or use of claim 26, wherein the non-ionic surfactant is a polysorbate surfactant, such as polysorbate 80 or polysorbate 20.

29. The aqueous solution, method or use of claim 26, wherein the non-ionic surfactant is an alkyl ether of polyethylene glycol.

30. The aqueous solution, method or use of claim 29, wherein the alkyl ether of polyethylene glycol is selected from polyethylene glycol (2) dodecyl ether, polyethylene glycol (2) oleyl ether and polyethylene glycol (2) hexadecyl ether.

31. The aqueous solution, method or use of claim 26, wherein the non-ionic surfactant is a block copolymer of polyethylene glycol and polypropylene glycol, such as poloxamer 188, poloxamer 407, poloxamer 171 or poloxamer 185.

32. The aqueous solution, method or use of claim 26, wherein the non-ionic surfactant is an alkylphenyl ether of polyethylene glycol, such as 4-(1,1,3,3-tetramethylbutyl)phenyl-polyethylene glycol.

33. The aqueous solution, method or use of any one of claims 26 to 32, wherein the non-ionic surfactant is present at a concentration of about 10 µg/mL to about 2000 µg/mL, such as about 50 µg/mL to about 1000 µg/mL, e.g. about 100 µg/mL to about 500 µg/mL.

34. The aqueous solution, method or use of any one of claims 1 to 33, further comprising an uncharged tonicity modifier, such as sucrose, trehalose, mannitol, sorbitol, PEG300 or PEG400.

35. The aqueous solution, method or use of claim 34, wherein the uncharged tonicity modifier is present at a concentration of about 50 mM to about 1000 mM, such as about 100 mM to about 500 mM, e.g. about 300 mM.

36. The aqueous solution, method or use of any one of claims 1 to 35, further comprising a charged tonicity modifier, such as selected from the group consisting of sodium chloride, sodium sulphate, sodium acetate, sodium lactate, glycine, histidine and arginine.

37. The aqueous solution, method or use of claim 36, wherein the charged tonicity modifier is present at a concentration of about 25 mM to about 500 mM, such as about 50 mM to about 250 mM, e.g. about 150 mM.

38. The aqueous solution, method or use of any one of claims 1 to 37, wherein the aqueous solution is isotonic.

39. The aqueous solution, method or use of any of claims 1 to 38, further comprising a preservative.

40. The aqueous solution, method or use of claim 39, wherein the preservative is selected from the group consisting of phenol, m-cresol, chlorocresol, benzyl alcohol, propylparaben, methylparaben, benzalkonium chloride and benzethonium chloride.

41. The aqueous solution, method or use of claim 39 or claim 40, wherein the preservative is present at a concentration of about 0.01 mM to about 100 mM.

42. The method of claim 2, or of any one of claims 4 to 41, wherein the method for stabilizing the antibody protein is a method for inhibiting formation of high molecular weight species of the antibody protein during storage.

43. The method of claim 2, or of any one of claims 4 to 41, wherein the method of stabilizing the antibody protein is a method for inhibiting formation of related species of the antibody protein during storage.

44. The method of claim 2, or of any one of claims 4 to 41, wherein the method of stabilizing the antibody protein is a method for inhibiting deamidation of the antibody protein during storage.

45. The method of claim 2, or of any one of claims 4 to 41, wherein the method of stabilizing the antibody protein is a method for inhibiting formation of low molecular weight degradation products in the aqueous solution during storage.

46. The method of claim 2, or of any one of claims 4 to 41, wherein the method of stabilizing the antibody protein is a method for inhibiting formation of visible particles in a composition of the antibody protein during storage.

47. The use of any one of claims 3 to 41, for inhibiting formation of high molecular weight species of the antibody protein during storage.

48. The use of any one of claims 3 to 41, for inhibiting formation of related species of the antibody protein during storage.

49. The use of any one of claims 3 to 41, for inhibiting deamidation of the antibody protein during storage.

50. The use of any one of claims 3 to 41, for inhibiting formation of low molecular weight degradation products of the antibody protein during storage.

51. The use of any one of claims 3 to 41, for inhibiting formation of visible particles in a composition of the antibody protein during storage.

52. The aqueous solution of claim 1 or any one of claims 4 to 41, wherein the solution is for administration by subcutaneous or intramuscular injection or by intravenous injection or infusion.

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2018/050481

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K9/08 A61K9/00 A61K38/17 A61K39/395
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, WPI Data, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2013/209465 A1 (JEZEK JAN [GB] ET AL) 15 August 2013 (2013-08-15)	1-7, 10-12, 17,18, 21-25, 36-39, 41-43, 45-48, 50-52
Y	example 3	1-15, 17-52
A		16

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[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

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Date of the actual completion of the international search	Date of mailing of the international search report
22 May 2018	01/06/2018

Name and mailing address of the ISA/	Authorized officer
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Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2008/112953 A1 (MCAULEY ARNOLD [US] ET AL) 15 May 2008 (2008-05-15)	1-3,6,8, 10-15, 21-26, 28,33, 36-38, 42-52
Y	claims 24, 48-55, 59-61, 64-70 paragraphs [0167] - [0181]	1-15, 17-52
A	figures 5-7	16

X	US 8 241 632 B2 (REHDER DOUGLAS [US] ET AL) 14 August 2012 (2012-08-14)	1-3,6,8, 10-15, 21-26, 28,33, 36-38, 42-52
Y	claims 1-9 page 35, paragraph 24-64	1-15, 17-52
A	figures 4-6	16

X	US 2003/138417 A1 (KAISHEVA ELIZABET A [US] ET AL) 24 July 2003 (2003-07-24)	1-3,6,8, 10-14, 21-26, 28,33, 36-38, 42-52
Y	claims 1, 3-9 example 3	1-15, 17-52
A		16

X,P	WO 2018/011404 A1 (PHILOGEN SPA [IT]) 18 January 2018 (2018-01-18)	1-8, 10-14, 21-26, 28, 34-38, 42-52
Y,P	claims 1-7, 14-18, 34-36 page 4, lines 26-35	1-15, 17-52
A,P	page 5, lines 16-22 examples 1, 2, 9	16

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/GB2018/050481

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2013209465 A1	15-08-2013	EP 2598167 A1	05-06-2013
		US 2013209465 A1	15-08-2013
		WO 2012013980 A1	02-02-2012
US 2008112953 A1	15-05-2008	AR 063150 A1	30-12-2008
		AU 2007307107 A1	17-04-2008
		CA 2665567 A1	17-04-2008
		CL 2007002880 A1	09-05-2008
		EP 2081553 A2	29-07-2009
		JP 5631591 B2	26-11-2014
		JP 2010505852 A	25-02-2010
		PE 11792008 A1	29-09-2008
		TW 200831129 A	01-08-2008
		US 2008112953 A1	15-05-2008
		WO 2008045373 A2	17-04-2008
US 8241632 B2	14-08-2012	AR 063149 A1	30-12-2008
		AU 2007309616 A1	02-05-2008
		CA 2666492 A1	02-05-2008
		CL 2007002881 A1	09-05-2008
		EP 2094247 A2	02-09-2009
		JP 5623743 B2	12-11-2014
		JP 2010506911 A	04-03-2010
		PE 08572008 A1	19-08-2008
		TW 200833357 A	16-08-2008
		US 2008124326 A1	29-05-2008
		US 2010158908 A1	24-06-2010
		WO 2008051363 A2	02-05-2008
US 2003138417 A1	24-07-2003	AT 556591 T	15-05-2012
		CA 2466034 A1	15-05-2003
		CN 1612689 A	04-05-2005
		DK 1441589 T3	06-08-2012
		EP 1441589 A2	04-08-2004
		ES 2392073 T3	04-12-2012
		HK 1074750 A1	08-06-2007
		IL 161677 A	16-06-2010
		JP 5290489 B2	18-09-2013
		JP 2005508981 A	07-04-2005
		JP 2011068675 A	07-04-2011
		KR 20050044365 A	12-05-2005
		NZ 532896 A	31-08-2007
		PT 1441589 E	13-08-2012
		US 2003138417 A1	24-07-2003
		US 2011070231 A1	24-03-2011
		US 2011318343 A1	29-12-2011
		WO 03039485 A2	15-05-2003
WO 2018011404 A1	18-01-2018	NONE	