



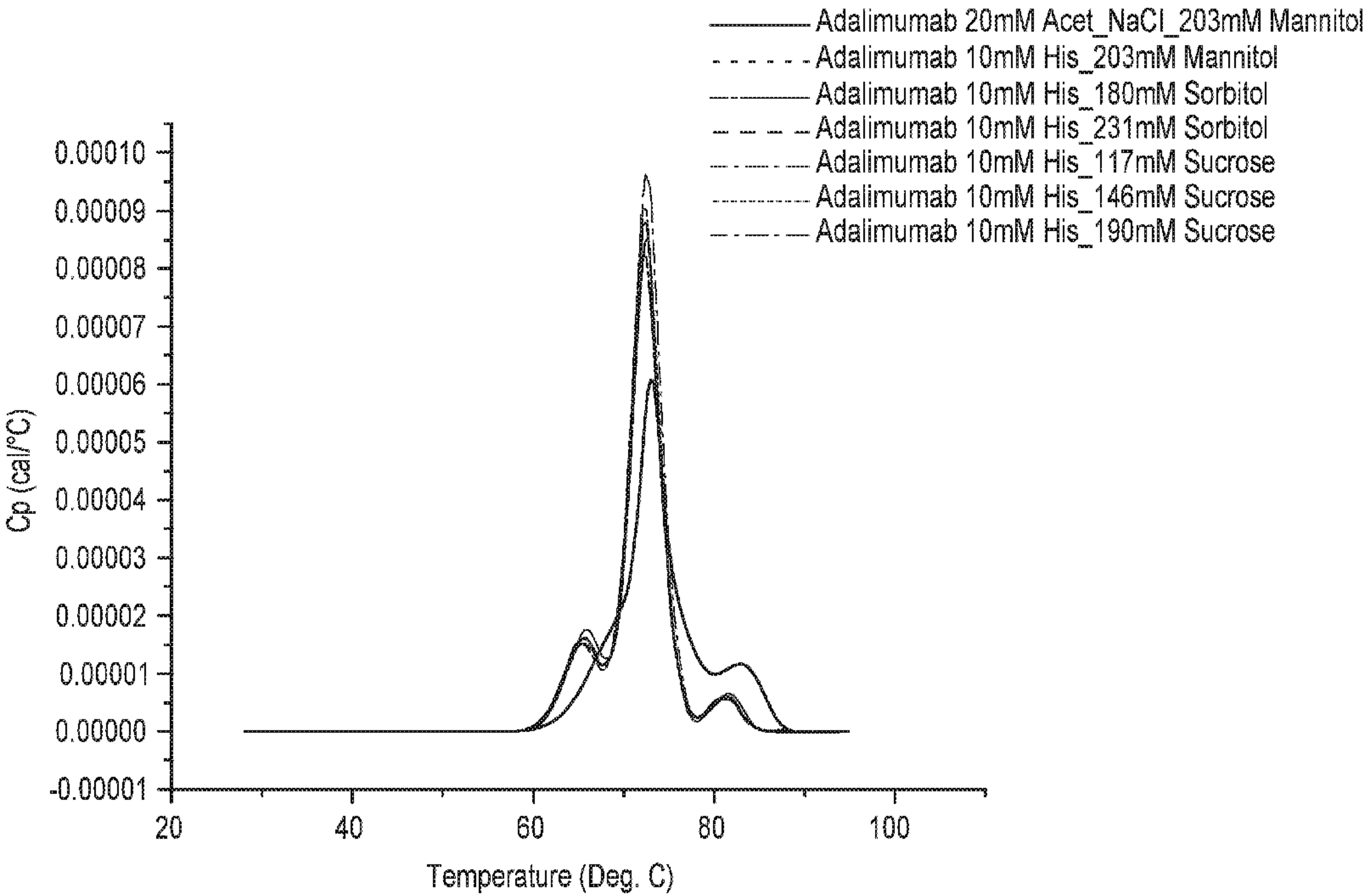
(12) **DEMANDE DE BREVET CANADIEN
CANADIAN PATENT APPLICATION**
(13) **A1**

(86) Date de dépôt PCT/PCT Filing Date: 2017/02/01
(87) Date publication PCT/PCT Publication Date: 2017/08/10
(85) Entrée phase nationale/National Entry: 2018/07/31
(86) N° demande PCT/PCT Application No.: US 2017/016040
(87) N° publication PCT/PCT Publication No.: 2017/136433
(30) Priorité/Priority: 2016/02/03 (US62/290,654)

(51) Cl.Int./Int.Cl. *C07K 16/24* (2006.01),
A61K 39/395 (2006.01)
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(54) Titre : FORMULATIONS DE TAMPON POUR AMELIORER LA STABILITE D'ANTICORPS
(54) Title: BUFFER FORMULATIONS FOR ENHANCED ANTIBODY STABILITY

FIGURE 16



(57) **Abrégé/Abstract:**
The disclosure provides buffered formulations of adalimumab. The formulations comprise a buffer comprising an acetate salt, sorbitol, histidine and/or a histidine salt and optionally arginine and/or an arginine salt, and polysorbate 80. The formulations have an acidic pH, and enhance the thermal, conformational and colloidal stability of antibodies, including the adalimumab antibody.

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property
Organization
International Bureau



WIPO | PCT



(10) International Publication Number
WO 2017/136433 A1

(43) International Publication Date
10 August 2017 (10.08.2017)

(51) International Patent Classification:

C07K 16/24 (2006.01) **A61K 39/395** (2006.01)

(21) International Application Number:

PCT/US2017/016040

(22) International Filing Date:

1 February 2017 (01.02.2017)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

62/290,654 3 February 2016 (03.02.2016) US

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(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

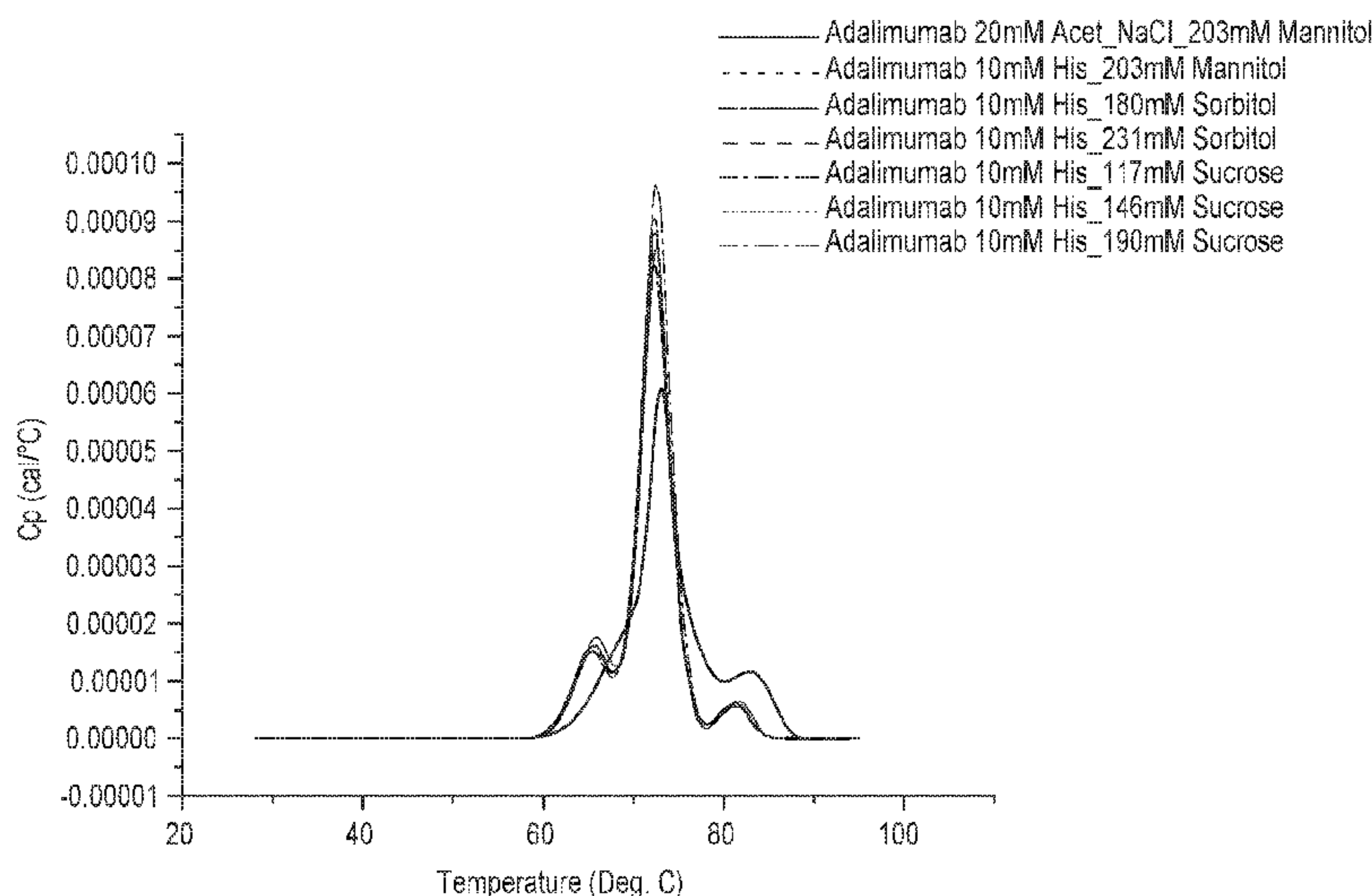
(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:

- with international search report (Art. 21(3))
- with sequence listing part of description (Rule 5.2(a))

(54) Title: BUFFER FORMULATIONS FOR ENHANCED ANTIBODY STABILITY

FIGURE 16



(57) Abstract: The disclosure provides buffered formulations of adalimumab. The formulations comprise a buffer comprising an acetate salt, sorbitol, histidine and/or a histidine salt and optionally arginine and/or an arginine salt, and polysorbate 80. The formulations have an acidic pH, and enhance the thermal, conformational and colloidal stability of antibodies, including the adalimumab antibody.

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BUFFER FORMULATIONS FOR ENHANCED ANTIBODY STABILITY

RELATED APPLICATIONS

[0001] This application claims priority to U.S. Patent Application No. 62/290,654, filed February 3, 2016, the contents of which are herein incorporated by reference in their entirety.

INCORPORATION OF SEQUENCE LISTING

[0002] The contents of the text file named “ONBI-006001WO_SeqList.txt”, which was created on January 30, 2017 and is 12 KB in size, are hereby incorporated by reference in their entirety.

FIELD OF THE DISCLOSURE

[0003] The disclosure relates generally to the field of antibody formulation chemistry. More particularly, the disclosure relates to buffered formulations for antibody storage, which enhance the thermal stability, conformational and colloidal stability of the antibody, thereby enhancing long term storage of the antibody.

BACKGROUND

[0004] Various publications, including patents, published applications, accession numbers, technical articles and scholarly articles are cited throughout the specification. Each of these cited publications is incorporated by reference herein, in its entirety and for all purposes.

[0005] As part of the Biologics Price Competition and Innovation Act (BPCIA), a biological drug product (produced in or derived from living organisms) may be demonstrated to be “biosimilar” if data show that, among other things, the product is “highly similar” to an approved biological product. The biosimilar product should retain at least the biologic function and treatment efficacy of the U.S. Food and Drug Agency-approved biological product. The biosimilar product may be formulated differently, however, from the approved biological product. The formulation may improve stability and shelf storage of the biologic drug product, and may also improve the efficacy in treating a particular disease or condition. The formulation may also improve other aspects of administration, including a reduction in patient discomfort or other untoward effects that a patient may experience upon administration of the approved biological product.

[0006] Antibody molecules may be used as biological drugs, and many such antibodies are approved for use in human beings. Antibody molecules may be produced as a biosimilar, and reformulated accordingly. There remains a need in the art for high-quality antibody biosimilars.

SUMMARY

[0007] The disclosure features buffered antibody formulations (*i.e.*, compositions of the disclosure), comprising (a) an antibody. The antibody may specifically bind to tumor necrosis factor alpha. The antibody may comprise a heavy chain comprising the amino acid sequence EVQLVESGGGLVQPGRSLRLSCAASGFTFDDYAMHWVRQAPGKGLEWVSAITWNSGHIDYADSVEGRFTISRDNANKNSLYLQMNSLRAEDTAVYYCAKVSYLSTASSLDYWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCDKTHTCTPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK (SEQ ID NO: 1) and a light chain comprising the amino acid sequence DIQMTQSPSSLSASVGDRVTITCRASQGIRNYLAWYQQKPGKAPKLLIYAASLTQSGVPSRFSGSGSGTDFTLTISSLQPEDVATYYCQRYNRAPYTFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO: 2).

[0008] In certain embodiments, the heavy chain of the antibody comprises a variable region comprising the amino acid sequence EVQLVESGGGLVQPGRSLRLSCAASGFTFDDYAMHWVRQAPGKGLEWVSAITWNSGHIDYADSVEGRFTISRDNANKNSLYLQMNSLRAEDTAVYYCAKVSYLSTASSLDYWGQGTLVTVSS (SEQ ID NO: 3), a first constant domain (CH1 domain) comprising the amino acid sequence ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKKV (SEQ ID NO: 4), a hinge region comprising the amino acid sequence EPKSCDKTHTCP (SEQ ID NO: 5), a second constant domain (CH2 domain) comprising the amino acid sequence PCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYK (SEQ ID NO: 6), and a third constant domain (CH3 domain) comprising the amino

acid sequence CKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCL VKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFS CSVMHEALHNHYTQKSLSLSPGK (SEQ ID NO: 7).

[0009] In certain embodiments, the light chain of the antibody comprises a variable region comprising the amino acid sequence DIQMTQSPSSLSASVGDRVTITCRASQGIRNYL AWYQQKPGKAPKLLIYAASLTQSGVPSRFSGSGSGTDFTLTISSLQPEDVATYYCQR YNRAPYTFGQGTKVEIKR (SEQ ID NO: 8) and a constant region comprising the amino acid sequence TVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQ SGNSQESVTEQDSKSTYSLSSLTLSKADYEEKHKVYACEVTHQGLSSPVTKSFNRG EC (SEQ ID NO: 9).

[0010] In certain embodiments, the antibody may comprise a variable heavy chain comprising the amino acid sequence EVQLVESGGGLVQPGRSLRLSCAASGFTFDDYAM HWVRQAPGKGLEWVSAITWNSGHIDYADSVEGRFTISRDNKNSLYLQMNSLRAED TAVYYCAKVSYLSTASSLDYWGGQGLVTVSS (SEQ ID NO: 3) and a variable light chain comprising the amino acid sequence DIQMTQSPSSLSASVGDRVTITCRASQGIR NYLAWYQQKPGKAPKLLIYAASLTQSGVPSRFSGSGSGTDFTLTISSLQPEDVATYYC QRYNRAPYTFGQGTKVEIKR (SEQ ID NO: 8).

[0011] The formulation (i.e., composition of the disclosure), in addition to the antibody, comprises (b) an aqueous buffer comprising from about 10 mM to about 30 mM of acetate or an acetate salt, preferably sodium acetate trihydrate, from about 15 mM to about 20 mM of histidine and/or a histidine salt and from about 0 mM to about 30 mM of arginine, from about 200 mM to about 206 mM of sorbitol, and (c) about 0.07% (v/v) to about 0.15% (v/v) of a non-ionic surfactant such as polysorbate 80. The buffered antibody formulation has a pH of from about 5.1 to about 5.3, preferably about 5.2.

[0012] The disclosure provides a buffered antibody formulation, comprising (a) an antibody comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2; (b) a buffer comprising (1) from about 1 mM to about 30 mM of an acetate salt, (2) from about 10 mM to about 30 mM of histidine and/or a histidine salt, (3) about 201 mM to about 205 mM of sorbitol, and (c) about 0.08% (v/v) to about 0.12% (v/v) of polysorbate 80, wherein the antibody formulation has a pH of from about 5.1 to about 5.3. In certain embodiments, the buffer of (b) further comprises from about 0.1 to about 30 mM of arginine and/or an arginine salt. In certain embodiments, the acetate salt comprises sodium acetate trihydrate. In certain embodiments,

the wherein the pH is about 5.2. In certain embodiments, the wherein the pH is about 5.2. In certain embodiments, the formulation does not comprise NaCl, a citrate, or a phosphate.

[0013] In some aspects, the formulation comprises from about 30 mg to about 70 mg of the antibody. In some preferred aspects, the formulation comprises from about 35 mg to about 65 mg of the antibody. In some preferred aspects, the formulation comprises from about 47 mg to about 53 mg of the antibody. In some preferred aspects, the formulation comprises about 50 mg of the antibody.

[0014] The buffer may comprise from about 1 mM to about 30 mM of the acetate salt. In some embodiments, the buffer comprises from about 10 mM to about 30 mM of the acetate salt. In some embodiments, the buffer comprises from about 15 mM to about 25 mM of the acetate salt. In some embodiments, the buffer comprises from about 20 mM of the acetate salt. In some embodiments, the buffer comprises from about 1 mM of the acetate salt. In some embodiments, the acetate salt comprises sodium acetate trihydrate.

[0015] The buffer may comprise from about 0.8 mM to about 1.2 mM of sodium acetate trihydrate, or from about 0.9 mM to about 1.1 mM of sodium acetate trihydrate, or about 1 mM of sodium acetate trihydrate.

[0016] The buffer may comprise from about 10 mM to about 30 mM of histidine or a histidine salt. In certain embodiments, the buffer may comprise from about 15 mM to about 25 mM of histidine or a histidine salt. In certain embodiments, the buffer may comprise from about 20 mM of histidine or a histidine salt. In certain embodiments, the histidine salt comprises L-histidine monohydrochloride monohydrate.

[0017] The buffer may comprise from about 10 mM to about 30 mM of L-histidine and L-histidine monohydrochloride monohydrate or about from about 10 mM to about 30 mM of L-histidine and L-histidine monohydrochloride monohydrate.

[0018] The buffer may comprise from about 0.1 mM to about 30 mM of arginine or an arginine salt. In certain embodiments, the buffer may comprise from about 10 mM to about 30 mM of arginine or an arginine salt. In certain embodiments, the buffer may comprise from about 18 mM to about 20 mM of arginine or an arginine salt. The buffer may comprise from about 19 mM of arginine or an arginine salt.

[0019] The buffer may comprise from about 201 mM to about 205 mM of sorbitol, or from about 202 mM to about 204 mM of sorbitol, or about 203 mM of sorbitol. The buffer is preferably substantially free or free of sodium chloride.

[0020] The buffered antibody formulation includes a non-ionic surfactant, which preferably is polysorbate 80. In some aspects, the formulation comprises from about 0.08% (v/v) to about 0.12% (v/v) of polysorbate 80. In some aspects, the formulation comprises from about 0.09% (v/v) to about 0.11% (v/v) of polysorbate 80. In some aspects, the formulation comprises about 0.1% (v/v) of polysorbate 80.

[0021] In a detailed aspect, a buffered antibody formulation comprises (a) about 30 mg to about 70 mg of an antibody comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, (b) an aqueous buffer comprising about 1 mM of an acetate salt, preferably sodium acetate trihydrate, from about 10 mM to about 30 mM of histidine and from about 0 mM to about 30 mM of arginine, about 203 mM of sorbitol, and (c) about 0.1% (by volume) of polysorbate 80. The buffered antibody formulation has a pH of from about 5.1 to about 5.3, preferably about 5.2. In some preferred aspects, the formulation comprises from about 35 mg to about 45 mg of the antibody. In some preferred aspects, the formulation comprises from about 37 mg to about 43 mg of the antibody. In some preferred aspects, the formulation comprises about 40 mg of the antibody.

[0022] The buffered antibody formulations may be used as a medicament, and may be used in methods of treatment. For example, the buffered antibody formulations may be for use in the treatment of arthritis. In some aspects, the buffered antibody formulations may be for use in the treatment of Rheumatoid Arthritis, or Juvenile Idiopathic Arthritis, or Psoriatic Arthritis. In some aspects, the buffered antibody formulations may be for use in the treatment of Ankylosing Spondylitis. In some aspects, the buffered antibody formulations may be for use in the treatment of Crohn's Disease. In some aspects, the buffered antibody formulations may be for use in the treatment of Ulcerative Colitis. In some aspects, the buffered antibody formulations may be for use in the treatment of Plaque Psoriasis.

[0023] The methods of treatment include methods for treating arthritis, including Rheumatoid Arthritis, Juvenile Idiopathic Arthritis, and Psoriatic Arthritis. The methods of treatment also include methods for treating Ankylosing Spondylitis, methods for treating Crohn's Disease, methods for treating Plaque Psoriasis, and methods for treating Ulcerative Colitis.

[0024] In some aspects, methods of treatment comprise administering to an arthritis patient, including a Rheumatoid Arthritis, Juvenile Idiopathic Arthritis, or Psoriatic Arthritis, an amount of the buffered antibody formulations described or exemplified herein effective to

treat the arthritis in the patient. In some aspects, methods of treatment comprise administering to an Ankylosing Spondylitis patient an amount of the buffered antibody formulations described or exemplified herein effective to treat the Ankylosing Spondylitis in the patient. In some aspects, methods of treatment comprise administering to a Crohn's Disease patient an amount of the buffered antibody formulations described or exemplified herein effective to treat the Crohn's Disease in the patient. In some aspects, methods of treatment comprise administering to an Ulcerative Colitis patient an amount of the buffered antibody formulations described or exemplified herein effective to treat the Ulcerative Colitis in the patient. In some aspects, methods of treatment comprise administering to a Plaque Psoriasis patient an amount of the buffered antibody formulations described or exemplified herein effective to treat the Plaque Psoriasis in the patient. The buffered antibody formulations are preferably administered subcutaneously to the patient, for example, by subcutaneous injection. The patient preferably is a human being.

[0025] The disclosure also provides kits, which may be used, for example, in accordance with the methods of treatment. Thus, for example, the kits generally comprise any of the buffered antibody formulations described or exemplified herein and instructions for using the formulation in a method of treatment. The method of treatment may be a method for treating arthritis. The method of treatment may be a method for treating Rheumatoid Arthritis. The method of treatment may be a method for treating Juvenile Idiopathic Arthritis. The method of treatment may be a method for treating Psoriatic Arthritis. The method of treatment may be a method for treating Ankylosing Spondylitis. The method of treatment may be a method for treating Crohn's Disease. The method of treatment may be a method for treating Ulcerative Colitis. The method of treatment may be a method for treating Plaque Psoriasis. The kits may include a device for administering the antibody formulation to a patient. The device may comprise a syringe and a needle. The device may comprise a catheter.

BRIEF DESCRIPTION OF THE DRAWINGS

[0026] FIG. 1 is an SE-UPLC % high molecular weight species (HMWS) micrograph showing trends in aggregation in adalimumab formulations at 55 °C for 10 days;

[0027] FIG. 2 is an SE-UPLC % high molecular weight species (HMWS) micrograph showing trends in aggregation in adalimumab formulations at 37 °C for 28 days;

[0028] FIG. 3 is an SE-UPLC % high molecular weight species (HMWS) micrograph showing trends in aggregation in adalimumab formulations at 30 °C for 28 days;

- [0029] FIG. 4 is an SE-UPLC % high molecular weight species (HMWS) micrograph showing trends in aggregation in adalimumab formulations at 5 °C for 28 days;
- [0030] FIG. 5 is a cation exchange chromatography (CEX-HPLC) chromatogram showing the % total acid species of samples of conditions 1-4 after exposure to three freeze/thaw cycles at -20 °C;
- [0031] FIG. 6 is a cation exchange chromatography (CEX-HPLC) chromatogram showing the % total acid species of samples of conditions 1-4 after exposure to three freeze/thaw cycles at -80 °C;
- [0032] FIG. 7 is a cation exchange chromatography (CEX-HPLC) chromatogram showing the % total acid species of samples of conditions 1-4 after exposure shaking stress;
- [0033] FIG. 8 is a plot of total acidic species formed in adalimumab biosimilar formulations at 55°C over a 10 day period;
- [0034] FIG. 9 is a plot of total acidic species formed in adalimumab biosimilar formulations at 5°C over a 28 day period;
- [0035] FIG. 10 is a plot of total acidic species formed in adalimumab biosimilar formulations at 30°C over a 28 day period;
- [0036] FIG. 11 is a plot of total acidic species formed in adalimumab biosimilar formulations at 37°C over a 28 day period;
- [0037] FIG. 12 is a spectrograph showing the intrinsic fluorescence emission scan tryptophan (Excitation 295/ Emission 310) for adalimumab biosimilar formulations with alternative buffers;
- [0038] FIG. 13 is a plot of DLS RH versus concentration of diluted protein for evaluated buffer systems;
- [0039] FIG. 14 is a plot of diffusion coefficient versus protein concentration for evaluated buffer systems; and
- [0040] FIG 15 is a Differential Scanning Calorimetry (DSC) plot depicting the thermal stability and related unfolding (thermograms) for all formulation compositions.
- [0041] FIG. 16 is a DSC plot depicting a thermal stability profile of Adalimumab in 10 mM histidine buffer pH 5.2. Each composition employs a different type and concentration of sugar for stabilization.
- [0042] FIG. 17 is a graph depicting the effect of histidine buffer molarity on the colloidal stability of Adalimumab. Histidine buffer at pH 5.2 including mannitol as a stabilizer.

[0043] FIG. 18 is a graph depicting the effect of histidine buffer molarity on the colloidal stability of Adalimumab. Histidine buffer at pH 5.2 including sorbitol as a stabilizer.

[0044] FIG. 19 is a graph depicting the effect of histidine buffer molarity on the colloidal stability of Adalimumab. Histidine buffer at pH 5.2 including sucrose as a stabilizer.

[0045] FIG. 20 is a DSC plot depicting a thermal stability profile of Adalimumab in 10 mM histidine buffer having a pH ranging between 5.0 and 6.0.

[0046] FIG. 21 is a graph depicting the effect of histidine buffer pH range on the colloidal stability of Adalimumab. Histidine buffer having a pH ranging between 5.0 and 6.0.

[0047] FIG. 22 is a DSC plot depicting a thermal stability profile of Adalimumab in 10 mM histidine-acetate buffer in comparison to alternative buffers.

[0048] FIG. 23 is a graph depicting the colloidal stability of Adalimumab in a Histidine-acetate compared to a histidine buffer or an acetate buffer. All buffers include sorbitol as a stabilizer. The slope for line plots of all 3 buffer types is identical and the R^2 value of each plot is greater than or equal to about 0.9.

[0049] FIG. 24 is a graph depicting the colloidal stability of Adalimumab in a Histidine-acetate compared to a histidine buffer or an acetate buffer. All buffers include sucrose as a stabilizer. The slope for line plots of all 3 buffer types is identical and the R^2 value of each plot is greater than or equal to about 0.9.

DETAILED DESCRIPTION

[0050] Various terms relating to aspects of the present disclosure are used throughout the specification and claims. Such terms are to be given their ordinary meaning in the art, unless otherwise indicated. Other specifically defined terms are to be construed in a manner consistent with the definition provided herein.

[0051] As used herein, the singular forms “a,” “an,” and “the” include plural referents unless expressly stated otherwise.

[0052] As used herein, the terms “comprising,” “having,” and “including” encompass the more restrictive terms “consisting essentially of” and “consisting of.”

[0053] The terms subject and patient are used interchangeably, and include any animal. Subjects include mammals, including companion and farm mammals, as well as rodents, including mice, rabbits, and rats, and other rodents. Non-human primates preferred subjects. Human beings are highly preferred subjects.

[0054] It has been observed in accordance with the disclosure that formulations of ONS-3010, which specifically binds to tumor necrosis factor alpha, can be buffered with histidine (and optionally arginine) amino acids and an acetate, while minimizing sodium chloride, with the buffers enhancing the thermal and colloidal stability of the antibody, even more so than formulations of adalimumab currently approved for patient use. It was observed that there is a fine balance in establishing and maintaining an acidic pH of about 5.2 with the appropriate salts and buffer components. It was observed, for example, that high levels of salt may induce aggregation and degradation, which could be improved by lowering the salt level. Accordingly, the disclosure features buffered formulations for antibodies, which formulations include an aqueous carrier comprising buffer comprising histidine (and optionally arginine) amino acids and an acetate, as well as mannitol and a non-ionic surfactant, but with minimal sodium chloride.

[0055] In some preferred aspects, the antibody specifically binds to an epitope on tumor necrosis factor alpha, and the epitope may be linear or conformational. In some preferred aspects, the antibody comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1. In some preferred aspects, the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 2. Preferably, the antibody comprises a heavy chain constant domain and/or a light chain constant domain. In highly preferred aspects, the antibody comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, an example of which is ONS-3010. The heavy and light chain amino acid sequences of the antibody may comprise those of U.S. Pat. No. 6,090,382.

[0056] Preferably, the antibody is a full length antibody, comprising both variable and constant regions, although in some aspects, the antibody may comprise a derivative or fragment or portion of a full-length antibody that retains the antigen-binding specificity, and also preferably retains most or all of the affinity, of the full length antibody molecule. The antibody may comprise post-translational modifications (PTMs) or moieties, which may impact antibody activity or stability. The antibody may be methylated, acetylated, glycosylated, sulfated, phosphorylated, carboxylated, and/or amidated, and may comprise other moieties that are well known in the art. Common PTMs for ONS-3010 include N-glycosylation, C-terminal variants (*e.g.*, cleavage of lysine, proline amidation), N-terminal pyro-E formation, oxidation, isomerization, deamidation, succinimide formation, mannosylation, K98 glycation, and fragmentation. Moieties include any chemical group or

combinations of groups commonly found on immunoglobulin molecules in nature, or otherwise added to antibodies by recombinant expression systems, including prokaryotic and eukaryotic expression systems.

[0057] The formulation preferably comprises a therapeutically effective amount of an antibody. The antibody may be any antibody compatible with the aqueous buffer formulation. A preferred antibody comprises a heavy chain having the amino acid sequence of SEQ ID NO: 1 and a light chain having the amino acid sequence of SEQ ID NO: 2. A therapeutically effective amount may vary, depending on the disease or condition being treated upon administration of the antibody, and/or depending on the characteristics of the subject to which the antibody is administered, such as age, gender, height, weight, state of advancement or stage of the disease or condition, the number and efficacy of previous administrations, other therapeutic agents administered to the subject, and other characteristics that are known to the practitioner or that would otherwise be taken into account in determining appropriate dosing. Preferably, a therapeutically effective amount is an amount that is effective to treat Rheumatoid Arthritis. In some preferred aspects, a therapeutically effective amount is an amount that is effective to treat Juvenile Idiopathic Arthritis, Psoriatic Arthritis, Ankylosing Spondylitis, Crohn's Disease, Plaque Psoriasis, Ulcerative Colitis, Inflammatory Bowel Disease, Hidradenitis Suppurativa, or Refractory Asthma.

[0058] The formulation may comprise from about 10 mg to about 70 mg of the antibody. In some aspects, the formulation comprises from about 20 mg to about 60 mg of the antibody. In some aspects, the formulation comprises from about 30 mg to about 50 mg of the antibody. In some aspects, the formulation comprises from about 35 mg to about 45 mg of the antibody. In some aspects, the formulation comprises from about 37 mg to about 43 mg of the antibody. In some aspects, the formulation comprises from about 38 mg to about 42 mg of the antibody. In some aspects, the formulation comprises from about 39 mg to about 41 mg of the antibody. In some aspects, the formulation comprises from about 30 mg to about 60 mg of the antibody. In some aspects, the formulation comprises from about 35 mg to about 55 mg of the antibody. In some aspects, the formulation comprises from about 40 mg to about 60 mg of the antibody. These ranges include the lower and upper amounts that define the range. In some aspects, the formulation comprises about 40 mg of the antibody.

[0059] The antibody is preferably formulated with a buffered aqueous carrier, and the carrier preferably comprises water. The buffered antibody formulation is preferably in liquid form, and more preferably in liquid form suitable for subcutaneous administration. Thus, the

amount of water in the buffered formulation may vary in accordance with the desired volume of the injectable bolus. The buffer comprises sodium acetate trihydrate, histidine and/or a histidine salt (and optionally arginine and/or an arginine salt), mannitol, sodium chloride, and a non-ionic surfactant, and maintains the antibody formulation at an acidic pH. Prefer the buffer is a histidine acetate buffer. When stored in the buffered formulation, the antibody is shelf-stable under normal storage conditions.

[0060] The buffer may comprise from about 1.0 mM to about 30 mM of an acetate salt. In some aspects, the buffer may comprise from about 10 mM to about 30 mM of an acetate salt. In some aspects, the buffer may comprise from about 12 mM to about 28 mM of an acetate salt. In some aspects, the buffer may comprise from about 14 mM to about 26 mM of an acetate salt. In some aspects, the buffer may comprise from about 15 mM to about 25 mM of an acetate salt. In some aspects, the buffer may comprise from about 16 mM to about 24 mM of an acetate salt. In some aspects, the buffer may comprise from about 17 mM to about 23 mM of an acetate salt. In some aspects, the buffer may comprise from about 18 mM to about 22 mM of an acetate salt. In some aspects, the buffer may comprise from about 19 mM to about 21 mM of an acetate salt. These ranges include the lower and upper amounts that define the range. In some aspects, the buffer comprises about 20 mM of an acetate salt. The acetate salt may comprise any suitable acetate salt. Non-limiting examples of preferred acetate salts include magnesium acetate salts, potassium acetate salts, calcium acetate salts, zinc acetate salts, and sodium acetate salts. More preferred acetate salts include anhydrous sodium acetate and sodium acetate trihydrate. Sodium acetate trihydrate is highly preferred.

[0061] An acetate buffer can be prepared by acetic acid and sodium acetate trihydrate as exemplified in the table below:

20 mM Sodium Acetate			
	pH	Acetic Acid Glacial g/L	Sodium Acetate Trihydrate g/L
	4.50	0.75	1.03
	4.70	0.61	1.34
	5.00	0.40	1.80
	5.20	0.29	2.06
	5.50	0.16	2.35
	6.00	0.06	2.59
	6.50	0.02	2.68

[0062] The buffer may comprise from about 10 mM to about 30 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 12 mM to about 28 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 14 mM to about 26 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 15 mM to about 25 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 16 mM to about 24 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 17 mM to about 23 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 18 mM to about 22 mM of histidine and/or a histidine salt. In some aspects, the buffer may comprise from about 19 mM to about 21 mM of histidine and/or a histidine salt. These ranges include the lower and upper amounts that define the range. In some aspects, the buffer comprises about 20 mM of histidine. Preferably, the histidine is L-histidine. Non-limiting examples of preferred histidine salts include L-histidine monohydrochloride monohydrate, L-histidine citrate, L-histidine glutamate, L-histidine succinate, and L-histidine aspartate. L-histidine monohydrochloride monohydrate is highly preferred.

[0063] A histidine buffer can be prepared by mixing histidine base and histidine hydrochloride as indicated in the table below.

20 mM Histidine Hydrochloride			
	pH	Histidine base g/L	Histidine HCl g/L
	4.50	0.08	4.08
	4.70	0.13	4.02
	5.00	0.25	3.86
	5.20	0.38	3.68
	5.50	0.67	3.28
	6.00	1.47	2.21
	6.50	2.31	1.07

[0064] A histidine acetate buffer can be prepared by mixing L-histidine base and acetic acid as indicated in the table below.

20 mM Histidine-Acetate		
pH	Volume of 20mM L- Histidine base (mL)	Volume of 18mM Acetic Acid (mL)
7.50	10.00	0.00
5.00	10.00	14.50
5.50	10.00	9.50
6.00	10.00	6.00
6.50	10.00	3.00

[0065] The buffer optionally comprises from about 10 mM to about 30 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 12 mM to about 28 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 14 mM to about 26 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 15 mM to about 25 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 16 mM to about 24 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 17 mM to about 23 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 18 mM to about 22 mM of arginine and/or an arginine salt. In some aspects, the buffer may comprise from about 19 mM to about 21 mM of arginine and/or an arginine salt. These ranges include the lower and upper amounts that define the range. In some aspects, the buffer comprises about 20 mM of arginine and/or an arginine salt. Non-limiting examples of preferred arginine salts include arginine acetate, arginine glutamate, arginine succinate, and arginine aspartate. Arginine acetate is highly preferred.

[0066] The buffer may comprise from about 100 mM to about 300 mM of sorbitol. In some aspects, the buffer may comprise from about 110 mM to about 290 mM of sorbitol. In some aspects, the buffer may comprise from about 120 mM to about 280 mM of sorbitol. In some aspects, the buffer may comprise from about 150 mM to about 250 mM of sorbitol. In some aspects, the buffer may comprise from about 175 mM to about 225 mM of sorbitol. In some aspects, the buffer may comprise from about 180 mM to about 220 mM of sorbitol. In some aspects, the buffer may comprise from about 185 mM to about 215 mM of sorbitol. In some aspects, the buffer may comprise from about 190 mM to about 215 mM of sorbitol. In some

aspects, the buffer may comprise from about 195 mM to about 210 mM of sorbitol. In some aspects, the buffer may comprise from about 197 mM to about 209 mM of sorbitol. In some aspects, the buffer may comprise from about 198 mM to about 208 mM of sorbitol. In some aspects, the buffer may comprise from about 198 mM to about 205 mM of sorbitol. In some aspects, the buffer may comprise from about 199 mM to about 207 mM of sorbitol. In some aspects, the buffer may comprise from about 200 mM to about 210 mM of sorbitol. In some aspects, the buffer may comprise from about 200 mM to about 207 mM of sorbitol. In some aspects, the buffer may comprise from about 200 mM to about 206 mM of sorbitol. In some aspects, the buffer may comprise from about 200 mM to about 205 mM of sorbitol. In some aspects, the buffer may comprise from about 200 mM to about 203 mM of sorbitol. In some aspects, the buffer may comprise from about 201 mM to about 205 mM of sorbitol. In some aspects, the buffer may comprise from about 201 mM to about 204 mM of sorbitol. In some aspects, the buffer may comprise from about 201 mM to about 203 mM of sorbitol. In some aspects, the buffer may comprise from about 202 mM to about 204 mM of sorbitol. In some aspects, the buffer may comprise from about 202 mM to about 203 mM of sorbitol. In some aspects, the buffer may comprise from about 202 mM to about 206 mM of sorbitol. These ranges include the lower and upper amounts that define the range. In some aspects, the buffer comprises about 203 mM of sorbitol.

[0067] In preferred embodiments, the buffer is substantially free or free of sodium chloride, citrates, and phosphates.

[0068] The antibody formulation preferably comprises a non-ionic surfactant. More preferably, the non-ionic surfactant comprises polysorbate 80. The antibody formulation, including the antibody and the aqueous buffer, preferably comprises from about 0.01% to about 1% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.03% to about 0.7% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.05% to about 0.4% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.075% to about 0.3% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.07% to about 0.25% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.07% to about 0.2% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.07% to about 0.15% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.07% to about 0.14% (by volume) of polysorbate 80. In some

aspects, the antibody formulation comprises from about 0.08% to about 0.3% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.08% to about 0.2% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.08% to about 0.15% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.08% to about 0.12% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.08% to about 0.1% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.09% to about 0.15% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.09% to about 0.2% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.09% to about 0.18% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.09% to about 0.11% (by volume) of polysorbate 80. In some aspects, the antibody formulation comprises from about 0.09% to about 0.1% (by volume) of polysorbate 80. These ranges include the lower and upper amounts that define the range. In some aspects, the antibody formulation comprises about 0.1% (by volume) of polysorbate 80. [0069] Polysorbate 20 can also be used in the disclosed compositions instead of polysorbate 80.

[0070] The antibody formulation preferably is buffered to an acidic pH. The formulation preferably has a pH of about 4.8 to about 5.6. In some aspects, the formulation has a pH of about 4.9 to about 5.5. In some aspects, the formulation has a pH of about 5.0 to about 5.4. In some preferred aspects, the formulation has a pH of about 5.0 to about 5.3. In some preferred aspects, the formulation has a pH of about 5.0 to about 5.2. In some aspects, the formulation has a pH of about 5.1 to about 5.3. In some aspects, the formulation has a pH of about 5.1 to about 5.5. In some preferred aspects, the formulation has a pH of about 5.1 to about 5.2. In some preferred aspects, the formulation has a pH of about 5.1 to about 5.4. In some aspects, the formulation has a pH of about 5.2 to about 5.4. In some aspects, the formulation has a pH of about 5.2 to about 5.5. In some preferred aspects, the formulation has a pH of about 5.2 to about 5.3. These ranges include the lower and upper amounts that define the range. In some aspects, the formulation has a pH of about 5.2.

[0071] In some preferred aspects, the antibody formulation comprises about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer comprising about 0.7 mM to

about 1.3 mM of sodium acetate trihydrate, about 200 mM to about 206 mM of sorbitol, about 10 mM to about 30 mM of L-histidine and/or a histidine salt, 0 mM to about 30 mM of arginine and/or an arginine salt, and about 0.07% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists essentially of about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting essentially of about 0.7 mM to about 1.3 mM of sodium acetate trihydrate, about 200 mM to about 206 mM of sorbitol, about 12 mM to about 28 mM of L-histidine and/or a histidine salt, 0 mM to about 28 mM of arginine and/or an arginine salt, and about 0.07% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists of about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting of about 0.7 mM to about 1.3 mM of sodium acetate trihydrate, about 200 mM to about 206 mM of sorbitol, about 14 mM to about 26 mM of L-histidine and/or a histidine salt, 0 mM to about 26 mM of arginine and/or an arginine salt, and about 0.07% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In any such embodiments, the antibody may be present in the formulation at about 37 mg to about 43 mg, or about 38 mg to about 42 mg, or about 39 mg to about 41 mg, or about 40 mg.

[0072] In some preferred aspects, the antibody formulation comprises about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer comprising about 0.8 mM to about 1.2 mM of an acetate salt, about 201 mM to about 205 mM of sorbitol, about 15 mM to about 25 mM of L-histidine and/or a histidine salt, 0 mM to about 25 mM of arginine and/or an arginine salt, and about 0.08% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists essentially of about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting essentially of about 0.8 mM to about 1.2 mM of an acetate salt, about 201 mM to about 205

mM of sorbitol, about 16 mM to about 24 mM of L-histidine and/or a histidine salt, 0 mM to about 24 mM of arginine and/or an arginine salt, and about 0.08% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists of about 35 mg to about 45 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting of about 0.8 mM to about 1.2 mM of an acetate salt, about 201 mM to about 205 mM of sorbitol, about 17 mM to about 23 mM of L-histidine and/or a histidine salt, 0 mM to about 23 mM of arginine and/or an arginine salt, and about 0.08% to about 0.15% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In any such embodiments, the antibody may be present in the formulation at about 37 mg to about 43 mg, or about 38 mg to about 42 mg, or about 39 mg to about 41 mg, or about 40 mg. The acetate salt may comprise any suitable acetate salt. Non-limiting examples of preferred acetate salts include magnesium acetate salts, potassium acetate salts, calcium acetate salts, zinc acetate salts, and sodium acetate salts. More preferred acetate salts include anhydrous sodium acetate and sodium acetate trihydrate. Sodium acetate trihydrate is highly preferred.

[0073] In some preferred aspects, the antibody formulation comprises about 39 mg to about 41 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer comprising about 0.9 mM to about 1.1 mM of an acetate salt, about 202 mM to about 204 mM of sorbitol, about 18 mM to about 22 mM of L-histidine and/or a histidine salt, 0 mM to about 22 mM of arginine and/or an arginine salt, and about 0.09% to about 0.11% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists essentially of about 39 mg to about 41 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting essentially of about 0.9 mM to about 1.1 mM of an acetate salt, about 202 mM to about 204 mM of sorbitol, about 19 mM to about 21 mM of L-histidine and/or a histidine salt, 0 mM to about 21 mM of arginine and/or an arginine salt, and about 0.09% to about 0.11% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In some aspects, the antibody formulation consists of about 39 mg to about 41 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid

sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting of about 0.9 mM to about 1.1 mM of an acetate salt, about 202 mM to about 204 mM of sorbitol, about 20 mM of L-histidine and/or a histidine salt, 0 mM to about 20 mM of arginine and/or an arginine salt, and about 0.09% to about 0.11% (by volume) of polysorbate 80, and has a pH of about 5.1 to about 5.3. In any such embodiments, the antibody may be present in the formulation at about 37 mg to about 43 mg, or about 38 mg to about 42 mg, or about 39 mg to about 41 mg, or about 40 mg. The acetate salt may comprise any suitable acetate salt. Non-limiting examples of preferred acetate salts include magnesium acetate salts, potassium acetate salts, calcium acetate salts, zinc acetate salts, and sodium acetate salts. More preferred acetate salts include anhydrous sodium acetate and sodium acetate trihydrate. Sodium acetate trihydrate is highly preferred.

[0074] In some preferred aspects, the antibody formulation comprises about 40 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer comprising about 1 mM of an acetate salt, about 203 mM of sorbitol, about 10 mM to about 30 mM of L-histidine and/or a histidine salt, 0 mM to about 30 mM of arginine and/or an arginine salt, and about 0.1% (by volume) of polysorbate 80, and has a pH of about 5.2. In some aspects, the antibody formulation consists essentially of about 40 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting essentially of about 1 mM of an acetate salt, about 203 mM of sorbitol, about 12 mM to about 28 mM of L-histidine and/or a histidine salt, 0 mM to about 28 mM of arginine and/or an arginine salt, and about 0.1% (by volume) of polysorbate 80, and has a pH of about 5.2. In some aspects, the antibody formulation consists of about 40 mg of an antibody that specifically binds to tumor necrosis factor alpha and comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2, a buffer consisting of about 1 mM of an acetate salt, about 203 mM of sorbitol, about 14 mM to about 26 mM of L-histidine and/or a histidine salt, 0 mM to about 26 mM of arginine and/or an arginine salt, and about 26.35 mM of sodium chloride, and about 0.1% (by volume) of polysorbate 80, and has a pH of about 5.2. The acetate salt may comprise any suitable acetate salt. Non-limiting examples of preferred acetate salts include magnesium acetate salts, potassium acetate salts, calcium acetate salts, zinc acetate salts, and

sodium acetate salts. More preferred acetate salts include anhydrous sodium acetate and sodium acetate trihydrate. Sodium acetate trihydrate is highly preferred.

[0075] The formulation stabilizes the antibody for improved shelf storage, particularly over a period of months to years. When stored in the formulation, the antibody maintains thermal and colloidal stability during the period of storage. For example, when stored in the formulation, the antibody is stable and exhibits minimal aggregation, flocculation, fragmentation, and denaturation, and the antibody retains its tumor necrosis factor alpha binding activity.

[0076] It is preferred that the antibody formulation be stored under refrigerated conditions, and preferably at a temperature of from about 2° C to about 8° C, including from about 2° C to about 6° C, and including about 2° C, about 3° C, about 4° C, about 5° C, about 6° C, about 7° C, or about 8° C. The antibody formulation may be stored at such temperatures for at least about 3 months. In some aspects, the antibody formulation may be stored at such temperatures for at least about 6 months. In some aspects, the antibody formulation may be stored at such temperatures for at least about 9 months. In some aspects, the antibody formulation may be stored at such temperatures for at least about 12 months. In some aspects, the antibody formulation may be stored at such temperatures for at least about 15 months. In some aspects, the antibody formulation may be stored at such temperatures for at least about 18 months. During the storage period the antibody is stable and exhibits minimal aggregation, flocculation, fragmentation, and denaturation, and the antibody retains its tumor necrosis factor alpha binding activity such that the antibody formulation may be removed from storage, administered to a patient, and still exhibit therapeutic efficacy against the condition for which the formulation is administered.

[0077] The formulation comprises about 10 mg to about 70 mg of antibody. Among this amount of antibody protein is a percentage of antibody monomers in active, native form, as well as a percentage of antibody fragments, antibody aggregates, and denatured or partially denatured antibodies that have reduced or no tumor necrosis binding activity. It is highly preferred that the formulation include a maximal amount of functional antibody monomers and a minimal amount of antibody fragments, aggregates, and structurally altered forms of the antibody with reduced binding activity and/or therapeutic efficacy (relative to the unaltered monomer). For example, the antibody formulation preferably contains at least about 85% by weight of antibody monomers, and less than about 15% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor

alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months.

[0078] In some aspects, the antibody formulation contains at least about 90% by weight of antibody monomers, and less than about 10% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 93% by weight of antibody monomers, and less than about 7% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 95% by weight of antibody monomers, and less than about 5% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 96% by weight of antibody monomers, and less than about 4% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 97% by weight of antibody monomers, and less than about 3% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 98% by weight of antibody monomers, and less than about 2% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. In some aspects, the antibody formulation contains at least about 99% by weight of antibody monomers, and less than about 1% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about six months. The amount of antibody monomers and antibody fragments, aggregates, and structurally altered forms may be determined according to any technique suitable in the art, including those described or exemplified herein, including any one or combination of

dynamic light scattering (DLS), differential scanning calorimetry (DSC), size exclusion chromatography (SE-UPLC), non-reducing and reducing capillary electrophoresis SDS (NR CE-SDS and R CE-SDS), peptide mapping and particle counting (PC).

[0079] In some aspects, the antibody formulation contains at least about 90% by weight of antibody monomers, and less than about 10% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 93% by weight of antibody monomers, and less than about 7% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 95% by weight of antibody monomers, and less than about 5% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 96% by weight of antibody monomers, and less than about 4% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 97% by weight of antibody monomers, and less than about 3% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 98% by weight of antibody monomers, and less than about 2% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. In some aspects, the antibody formulation contains at least about 99% by weight of antibody monomers, and less than about 1% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about twelve months. The amount of antibody monomers and antibody fragments, aggregates, and structurally altered forms may be determined according to any technique suitable in the art,

including those described or exemplified herein, including any one or combination of dynamic light scattering (DLS), differential scanning calorimetry (DSC), size exclusion chromatography (SE-UPLC), non-reducing and reducing capillary electrophoresis SDS (NR CE-SDS and R CE-SDS), peptide mapping and particle counting (PC).

[0080] In some aspects, the antibody formulation contains at least about 90% by weight of antibody monomers, and less than about 10% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 93% by weight of antibody monomers, and less than about 7% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 95% by weight of antibody monomers, and less than about 5% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 96% by weight of antibody monomers, and less than about 4% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 97% by weight of antibody monomers, and less than about 3% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 98% by weight of antibody monomers, and less than about 2% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. In some aspects, the antibody formulation contains at least about 99% by weight of antibody monomers, and less than about 1% by weight of antibody fragments, aggregates, and structurally altered forms with reduced tumor necrosis factor alpha binding activity and/or therapeutic efficacy when stored at about 2° C to about 8° C for at least about eighteen months. The amount of antibody monomers and antibody fragments, aggregates,

and structurally altered forms may be determined according to any technique suitable in the art, including those described or exemplified herein, including any one or combination of dynamic light scattering (DLS), differential scanning calorimetry (DSC), size exclusion chromatography (SE-UPLC), non-reducing and reducing capillary electrophoresis SDS (NR CE-SDS and R CE-SDS), peptide mapping and particle counting (PC).

[0081] The disclosure also features methods for treating Rheumatoid Arthritis in a subject in need thereof by administering a therapeutically effective amount of any of the antibody formulations described or exemplified herein. The disclosure also features methods for treating Juvenile Idiopathic Arthritis, Psoriatic Arthritis, Ankylosing Spondylitis, Crohn's Disease, Plaque Psoriasis, Ulcerative Colitis, Inflammatory Bowel Disease, Hidradenitis Suppurativa, or Refractory Asthma by administering a therapeutically effective amount of any of the antibody formulations described or exemplified herein. Therapeutic efficacy is attained, for example, by the ONS-3010 antibody present in the administered formulation. Administration of the antibody formulation may be according to any suitable route, preferably by injection, and more preferably by subcutaneous injection. Administration may be carried out under the direction or supervision of a medical practitioner.

[0082] The antibody formulations described and exemplified herein may be for use as a medicament. The antibody formulations described and exemplified herein may be for use in the manufacture of a medicament. The formulations may be for use in the treatment of Rheumatoid Arthritis. The formulations may be for use in the treatment of Juvenile Idiopathic Arthritis. The formulations may be for use in the treatment of Psoriatic Arthritis. The formulations may be for use in the treatment of Ankylosing Spondylitis. The formulations may be for use in the treatment of Crohn's Disease. The formulations may be for use in the treatment of Plaque Psoriasis. The formulations may be for use in the treatment of Ulcerative Colitis. The formulations may be for use in the treatment of Inflammatory Bowel Disease. The formulations may be for use in the treatment of Hidradenitis Suppurativa. The formulations may be for use in the treatment of Refractory Asthma.

[0083] The disclosure also features kits. The kits may be used, for example, to practice any of the methods described or exemplified herein. In some aspects, a kit comprises any antibody formulation described or exemplified herein, and instructions for using the antibody formulation in any of the methods or uses described or exemplified herein. The kit may comprise a device for injecting the antibody formulation into a subject, including but not limited to a syringe and needle, or catheter.

[0084] The instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Rheumatoid Arthritis, including instructions for injecting the antibody formulation into a Rheumatoid Arthritis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Juvenile Idiopathic Arthritis, including instructions for injecting the antibody formulation into a Juvenile Idiopathic Arthritis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Psoriatic Arthritis, including instructions for injecting the antibody formulation into a Psoriatic Arthritis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Ankylosing Spondylitis, including instructions for injecting the antibody formulation into a Ankylosing Spondylitis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Crohn's Disease, including instructions for injecting the antibody formulation into a Crohn's Disease patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Plaque Psoriasis, including instructions for injecting the antibody formulation into a Plaque Psoriasis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Ulcerative Colitis, including instructions for injecting the antibody formulation into a Ulcerative Colitis patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Inflammatory Bowel Disease, including instructions for injecting the antibody formulation into an Inflammatory Bowel Disease patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Hidradenitis Suppurativa, including instructions for injecting the antibody formulation into a Hidradenitis Suppurativa patient in need thereof. In some aspects, the instructions included with the kit may include instructions for administering the antibody formulation in a method for treating Refractory Asthma, including instructions for injecting the antibody formulation into a Refractory Asthma patient in need thereof.

[0085] The following examples are provided to describe the disclosure in greater detail. They are intended to illustrate, not to limit, the disclosure.

Example 1

Materials and Methods

[0086] Introduction. Antibody ONS-3010 represents a biosimilar of adalimumab, and has been reformulated for enhanced storage stability. It is believed that the modifications to the buffer of the formulation composition may reduce the incidence of injection-site reaction, including injection pain and a burning sensation observed from subcutaneous administration of adalimumab (Kaiser C *et al.* (2012) *Rheumatol. Int.* 32:295–9, and Fransson J *et al.* (1996) *J. Pharm. Pharmacol.* 48:1012-5). Current adalimumab formulations include (in addition to the antibody), sodium chloride, monobasic sodium phosphate dihydrate, dibasic sodium phosphate dihydrate, sodium citrate, citric acid monohydrate, sorbitol, polysorbate 80, and sterile water for injection. The experimental approach described below included a series of development work to reformulate adalimumab for therapeutic administration. This work focused primarily on evaluating the stressed stability of differing buffer compositions. Four conditions were compared to a control of the adalimumab reference product buffer (per 0.8 ml: 40 mg adalimumab, 4.93 mg sodium chloride, 0.69 mg monobasic sodium phosphate dihydrate, 1.22 mg dibasic sodium phosphate dihydrate, 0.24 mg sodium citrate, 1.04 mg citric acid monohydrate, 9.6 mg mannitol, 0.8 mg polysorbate 80, and Q.S. sterile water for injection, pH 5.2).

[0087] Differential Scanning Calorimetry. The DSC thermograms was obtained using a MicroCal VP-Capillary DSC system (Malvern Instruments Ltd). Samples were diluted to 0.5mg/mL using matching formulation buffer as a reference and held for a 15 minute equilibration time at 25°C. The matching formulation buffer blank was subtracted from the samples. The scan rate used for the samples was 60°C/h with a range from 25-95°C. The filtering period used was 8 s and the active cell volume was 130µL. The data was analyzed with MicroCal VP-Capillary DSC Automated Analysis Software which utilizes Origin 7.0 software (OriginLab® Corporation, Northampton, MA).

[0088] Dynamic Light Scattering (DLS). The DLS testing method used a Wyatt DynaPro™ Plate Reader to provide information on protein size distribution and overall colloidal stability in solution. Hydrodynamic radius provided information on the presence of aggregation and confirmation of the molecule's structure in solution. DLS testing provided an orthogonal measure of size distribution in solution under non-denaturing conditions.

[0089] Size Exclusion Chromatography (SE-UPLC). SE-UPLC was used to monitor ONS-3010 size variant distribution. The SE-UPLC testing method separates proteins based on size. The method is isocratic with a sodium phosphate running buffer, using a Waters Acquity UPLC BEH200 SEC column (1.7 μ m, 4.6 x 150 mm). Peaks were monitored using absorbance at 280 nm. Species eluting before the monomer peak were aggregates (HMWS) and peaks eluting after the monomer peak were degradants (LMWS).

[0090] Non-Reducing and Reducing Capillary Electrophoresis SDS (NR CE-SDS and R CE-SDS). CE-SDS analysis was used to compare ONS-3010 size variants under denaturing conditions, with both non-reducing and reducing conditions, using a Beckman PA800 plus instrument. Capillary gel electrophoresis provides automated analysis of reduced and non-reduced proteins by size to determine protein purity and/or heterogeneity. Samples were treated with either an alkylation or reducing agent and SDS was bound to all proteins via a sample buffer. A polymer matrix was filled into the capillary prior to sample analysis. Samples were electrokinetically introduced to the capillary by an applied voltage, then electrophoresis was performed by applying a constant voltage to the capillary. The SDS treated proteins have mass to charge properties that are proportional to the protein weights, which allows for the separation of the SDS-bound proteins by the differences in molecular weight. Test article proteins were quantified by UV detection at 220 nm.

[0091] Cation Exchange Chromatography (CEX). For CEX analysis, a Dionex ProPac WCX-10 (4 x 250 mm) column was used. Mobile Phase A consisted of 2.4 mM Tris, 1.5 mM Imidazole, 11.6 mM Piperazine, pH 7.0 and mobile Phase B of 9.6 mM Tris, 6.0 mM Imidazole, 11.6 mM Piperazine, pH 11.0. The column temperature was maintained at 30 °C and the chromatogram was monitored and processed at 280 nm wavelength. Protein was eluted when applying a linear gradient from 10% B to 50% B in 23 mins at a flow rate of 0.8 mL/min.

[0092] Gradient info:

Time	% Mobile Phase A	% Mobile Phase B
0	90	10
20	90	10
25	50	50
30	50	50
30.5	20	80
35	20	80

35.5	90	10
45	90	10

[0093] Modulation of TNF-alpha activity: L929 Cell-Based Bioassay. The primary mechanism of action of adalimumab is the neutralization of circulating TNF-alpha. L929 cell-based bioassay measures cell death/viability. TNF-alpha induces cytotoxicity in L929 cells; relative potency of adalimumab was measured by monitoring live cells through a luminescent tag.

[0094] Peptide mapping. N-terminal sequence variants, C-terminal sequence variants, oxidation, deamidation, succinimide formation, isomerization are measured using peptide mapping LC-MS methodologies.

[0095] Particle count. The level of aggregates and particulates is a critical quality attribute to assess for liquid protein formulations. The presence of aggregates and particulates may negatively impact product quality.

Example 2

Results

[0096] This experimental series of studies focused on buffer composition, strength, and ability to achieve the desired pH of 5.2. Buffers tested included citrate and phosphate (which are used in the reference product formulation) and histidine acetate. Sodium chloride and mannitol concentrations (equivalent to those in adalimumab reference formulation) were added to conditions throughout experimental series 1 experiments. The following conditions/formulations were evaluated for stability of the adalimumab active.

1. Adalimumab Citrate-Phosphate formulation (Commercial Humira® formulation)

105.45 mM Sodium Chloride

5.53 mM Sodium Phosphate, Monobasic Dihydrate

8.57 mM Sodium Phosphate, Dibasic Dihydrate

1.02 mM Sodium Citrate, Dihydrate

6.19 mM Citric Acid, Monohydrate

65.87 mM Mannitol

0.1 % Polysorbate-80

pH 5.2

2. Adalimumab Acetate Salt Formulation

26.35 mM Sodium Chloride

20 mM Acetate

203.00 mM Mannitol

0.1% Polysorbate-80

pH 5.2

3. Adalimumab Histidine formulation

4.1% Sorbitol

5 mM L-histidine and L-histidine monohydrochloride monohydrate

0.015% Polysorbate 80

pH 5.5

4. Adalimumab Acetate Formulation

17mM acetate

4.7% sorbitol

0.01% polysorbate 20

pH 5.2

5. Adalimumab Histidine Acetate Formulation

20 mM Histidine Acetate

4% Mannitol (or 4% Sorbitol)

0.1% Polysorbate 80

pH 5.2

[0097] From this series of experiments, it was observed that some buffers were better than others at achieving and maintaining stability of the adalimumab active. The following is a summary of the results with reference to the FIGS.

[0098] Referring to FIGS. 1–4, SE-UPLC was used to measure and predict the long term colloidal stability of the adalimumab formulations of certain of the conditions 1-4 by looking at the colloidal behavior at various temperature ranges. As shown in Figure 1, 10 days at an elevated temperature of 55 °C causes significant aggregation (increasing SE-UPLC high molecular weight species or HMWS) is observed for condition 1 (Humira®) with a substantially lesser amount of aggregation for the conditions 3 and 4, respectively. Both conditions 3 and 4 were free of added NaCl. Such formulations lacking NaCl exhibited improved stability toward aggregation.

[0099] FIGS. 2-4 illustrate the aggregation observed over a 28 day period (real time) for conditions 1-4. As the temperature is lowered, the overall percentage of agglomeration of adalimumab (% HMWS as measured by SE-UPLC) decreases. It is evident that conditions 3 and 4 are more stable through a range of temperatures including 37 °C, 30 °C, and 5 °C for a period of 28 days in solution.

[00100] Freeze-thaw cycling was conducted for samples in the candidate formulations at two temperatures: -20° C and -80° C. Samples were placed in freezers set to the appropriate temperature and allowed to freeze thoroughly (for at least one hour). Samples were then removed from the freezer and allowed to thaw at 25° C (approximately 1 hour). This freezing step plus the thawing step constituted a single cycle. Samples were subjected to up to 3 freeze-thaw cycles, and then analyzed together by SE-UPLC, with a subset of samples also tested by NR CE-SDS.

[00101] FIGS. 5-6 illustrate the results of cation exchange chromatography (CEX-HPLC) of samples of conditions 1-4 after exposure to three freeze/thaw cycles at -20 °C (FIG. 5) and -80 °C (FIG. 6). This test provides a broad view of many physicochemical changes that can manifest themselves as changes in molecular charge. This includes specific charge based modifications such as deamidation, isomerization, and pyroglutamine formation. CEX-HPLC profiles were monitored for time zero through 3 freeze-thaw cycles, and the results were illustrated in FIGS. 5 and 6.

[00102] In order to assess the protective ability of the formulation toward shear forces, a shaking study was conducted in glass vials (1 mL fills) placed in an orbital shaking incubator set to 150 rpm at 25 °C. FIG. 7 illustrates the results of cation exchange chromatography (CEX-HPLC) of samples of conditions 1-4 after continuous shaking stress at 25 °C. This test provides a broad view of many physicochemical changes that can manifest themselves as changes in molecular charge. This includes specific charge based modifications such as deamidation, isomerization, and pyroglutamine formation. CEX-HPLC profiles were monitored for time zero through day 28.

[00103] To probe the behavior of the test formulations toward a stressed condition of elevated temperature, samples were incubated at 55° C for up to 10 days, and then tested by multiple analytical methods. While 55° C is well above storage conditions and any expected short-term handling conditions that would be encountered in the clinic, the stressed stability arm is extremely useful at highlighting formulation ability to protect from a myriad of forced degradation events that dominate at higher temperature.

[00104] FIG. 8 illustrates the results of cation exchange chromatography (CEX-HPLC) of samples of conditions 1-4 treated at 55 °C. This test provides a broad view of many physicochemical changes that can manifest themselves as changes in molecular charge. This includes specific charge based modifications such as deamidation, isomerization, and pyroglutamine formation, but can also reveal more subtle conformational shifts that can begin to occur at elevated temperatures. CEX-HPLC profiles were monitored for time zero through day 10.

[00105] FIGS. 9-11 illustrates the results of cation exchange chromatography (CEX-HPLC) of samples of conditions 1-4 treated at 5 °C, 30 °C, and 37 °C, respectively, in terms of acidic specie formation. This test provides a broad view of many physicochemical changes that can manifest themselves as changes in molecular charge. This includes specific charge based modifications such as deamidation, isomerization, and pyroglutamine formation, but can also reveal more subtle conformational shifts that can begin to occur at elevated temperatures. CEX-HPLC profiles were monitored for time zero through day 28. Here again, the compositions are all most stable at lower temperatures and more acidic species develop over time at higher temperatures. At such higher temperatures, there does not seem to be much difference when comparing the 4 conditions tested.

[00106] FIG. 12 is an emission scan showing intrinsic fluorescence emission of the formulations of conditions 2-5. While the fluorescence intensity varies for all the compositions, the wavelength maximum is similar indicating identical overall microenvironment for all tryptophan and tyrosine residues in the antibody.

[00107] Referring to FIG. 13, dynamic light scattering (DLS) was used to monitor the hydrodynamic radius R_h (size) of protein molecules in solution versus the concentration of diluted protein in solution. Hydrodynamic radius size in the 5 – 6 nanometer range under lower (~ 1mg/mL) protein concentration are typical for monomeric monoclonal antibodies (about 140 kDa in size); this size increases with protein concentration for condition 1, possibly due to crowding, self-association, or aggregation. Such higher sizes should typically be avoided under formulation conditions since they are indicative of an inherently unstable condition. Conditions 3 and 4, however, show significantly less agglomeration as the protein concentration increases.

[00108] FIG. 14 correlates well to that illustrated in FIG. 13 in that the agglomeration promoting conditions (e.g., Condition 1) have poor diffusivity compared to the conditions having smaller particles as the protein concentration increases.

[00109] Results based on stressed and accelerated stability studies indicate promising reformulation conditions with comparable and/or improved degradation rates relative to that of the antibody in the adalimumab reference formulation.

[00110] FIG. 15 shows the thermal stability and related unfolding (thermograms) for all formulation compositions as measured by Differential Scanning Calorimetry (DSC). The T_{onset} and T_m for the protein under all conditions besides Condition 1 is higher than that for Condition 1.

[00111] The invention is not limited to the embodiments described and exemplified above, but is capable of variation and modification within the scope of the appended claims.

Example 3

Results

[00112] This experimental series of studies focused on buffer composition, strength, and evaluation of the ability of histidine-acetate buffer pH of 5.2 to achieve conformational and colloidal stabilization of Adalimumab.

Conformational and Colloidal Stability of Adalimumab in Histidine buffer:

[00113] Conformational stability of Adalimumab in Histidine buffer pH 5.2 is evaluated by DSC. 3 types of sugars (mannitol, sorbitol, sucrose) are used as stabilizers along with Histidine buffer pH 5.2. The findings of the conformational stability trends are summarized in Figure 16. At lower pH of 5.2, an early unfolding event is observed which in case of IgG1 antibody is the C_H2 domain. Data from conformational study suggest that Adalimumab should be formulated in Histidine buffer at a $\text{pH} > 5.2$.

[00114] Colloidal stability of Adalimumab in Histidine buffer pH 5.2 is evaluated at varying molar concentrations of the buffer. 3 types of sugars (mannitol, sorbitol, sucrose) are used as stabilizers with these buffers. The findings of the colloidal stability trends are summarized in Figures 17, 18 and 19.

[00115] The colloidal stability for Adalimumab as depicted by an increase in average diffusion coefficient increases with decreasing buffer molarity. This trend is similar for the three types of sugars evaluated. Hence for better long term stability, lower molarity buffers would be considered ideal.

Effect of pH on Conformational and Colloidal stabilization of Adalimumab in Histidine buffer:

[00116] Conformational stability of Adalimumab in Histidine buffer is evaluated at varying pH in the range of pH 5 to 6. The findings of the conformational stability trends are

summarized in Figure 20. At lower pH (< 5.5), an early unfolding event is observed which in case of IgG1 antibody is the C_H2 domain. Data from conformational study suggest that Adalimumab should be formulated at a pH of 6.0 in Histidine buffer.

[00117] Colloidal stability of Adalimumab in Histidine buffer is evaluated at varying pH in the range of pH 5 to 6. The findings of the colloidal stability trends are summarized in Figure 21.

[00118] The colloidal stability for Adalimumab as depicted by an increase in average diffusion coefficient remains unchanged in pH range of 5 to 6.

Conformational and Colloidal Stability of Adalimumab in Histidine-Acetate buffer:

[00119] Conformational stability of Adalimumab in Histidine-Acetate buffer pH 5.2 is compared to that in Histidine buffer, Acetate buffer, citrate-phosphate buffer and water (buffer-less) composition. Two sugars (sucrose, sorbitol) are used as stabilizers with these buffers. The findings of the conformational stability trends are summarized in Figure 22.

[00120] The colloidal stability for Adalimumab in Histidine-Acetate buffer pH 5.2 is greater than that in Histidine buffer pH 5.2 or Acetate buffer pH 5.2 or citrate-phosphate buffer pH 5.2. Also as depicted by the melting temperature (T_m), the conformational stability of Adalimumab in Histidine-Acetate buffer pH 5.2 is equivalent to that in water (buffer-less) composition.

[00121] Colloidal stability of Adalimumab in Histidine-Acetate buffer pH 5.2 is compared to that in Histidine buffer and Acetate buffer. 2 sugars (mannitol, sorbitol) are used as stabilizers with these buffers. The findings of the colloidal stability trends are summarized in Figures 23 and 24. The colloidal stability for Adalimumab in Histidine-Acetate buffer pH 5.2 is equivalent to that in Histidine buffer pH 5.2 or Acetate buffer pH 5.2.

[00122] We claim:

CLAIMS

1. A buffered antibody formulation, comprising
 - (a) an antibody comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 1 and a light chain comprising the amino acid sequence of SEQ ID NO: 2;
 - (b) a buffer comprising
 - (1) from about 1 mM to about 30 mM of an acetate salt,
 - (2) from about 10 mM to about 30 mM of histidine and/or a histidine salt,
 - (3) about 201 mM to about 205 mM of sorbitol, and
 - (c) about 0.08% (v/v) to about 0.12% (v/v) of polysorbate 80,wherein the antibody formulation has a pH of from about 5.1 to about 5.3.
2. The buffered antibody formulation of claim 1, wherein the buffer of (b) further comprises from about 0.1 to about 30 mM of arginine and/or an arginine salt.
3. The buffered antibody formulation of claim 1 or 2, wherein the acetate salt comprises sodium acetate trihydrate.
4. The buffered antibody formulation of any one of claims 1-3, wherein the pH is about 5.2.
5. The buffered antibody formulation of any one of claims 1-4, wherein the formulation comprises from about 35 mg to about 45 mg of the antibody.
6. The buffered antibody formulation of any one of claims 1-4, wherein the formulation comprises from about 37 mg to about 43 mg of the antibody.
7. The buffered antibody formulation of any one of claims 1-4, wherein the formulation comprises about 40 mg of the antibody.
8. The buffered antibody formulation of any one of claims 1-7, wherein the formulation does not comprise NaCl, a citrate, or a phosphate.

9. The buffered antibody formulation of any one of claims 1-8, wherein the buffer comprises from about 10 mM to about 30 mM of the acetate salt.

10. The buffered antibody formulation of any one of claims 1-8, wherein the buffer comprises from about 15 mM to about 25 mM of the acetate salt.

11. The buffered antibody formulation of any one of claims 1-8, wherein the buffer comprises about 20 mM of the acetate salt.

12. The buffered antibody formulation of any one of claims 1-8, wherein the buffer comprises about 1 mM of the acetate salt.

13. The buffered antibody formulation of any of claims 1-12, wherein the buffer comprises from about 15 mM to about 25 mM of the histidine and/or a histidine salt.

14. The buffered antibody formulation of any one of claims 1-12, wherein the buffer comprises about 20 mM of the histidine and/or a histidine salt.

15. The buffered antibody formulation of any one of claims 1-14, wherein the buffer comprises from about 202 mM to about 204 mM of sorbitol.

16. The buffered antibody formulation of any one of claims 1-14, wherein the buffer comprises about 203 mM of sorbitol.

17. The buffered antibody formulation of any one of claims 2-16, wherein the buffer comprises from about 10 mM to about 30 mM of arginine and/or an arginine salt.

18. The buffered antibody formulation of any one of claims 2-16, wherein the buffer comprises from about 18 mM to about 20 mM of arginine and/or an arginine salt.

19. The buffered antibody formulation of any one of claims 2-16, wherein the buffer comprises about 19 mM of arginine and/or an arginine salt.

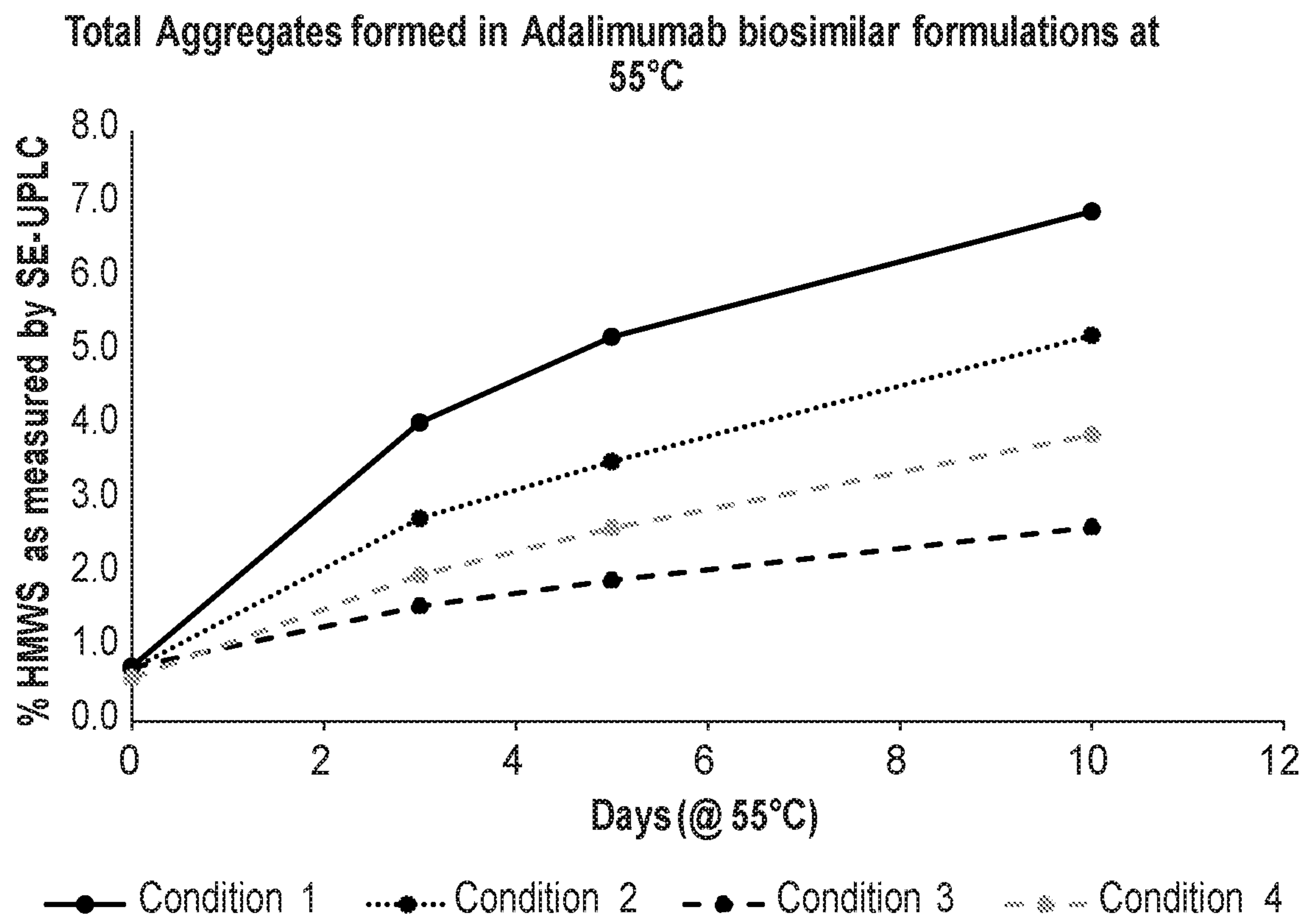
20. The buffered antibody formulation of any one of claims 1-19, wherein the formulation comprises from about 0.09% (v/v) to about 0.11% (v/v) of polysorbate 80.
21. The buffered antibody formulation of any one of claims 1-19, wherein the formulation comprises about 0.1% (v/v) of polysorbate 80.
22. A method for treating Rheumatoid Arthritis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Rheumatoid Arthritis.
23. The method of claim 22, wherein the subject is a human being.
24. The method of claim 22 or 23, wherein the administering step comprises subcutaneously injecting the antibody formulation.
25. A method for treating Juvenile Idiopathic Arthritis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Juvenile Idiopathic Arthritis.
26. The method of claim 25, wherein the subject is a human being.
27. The method of claim 25 or 26, wherein the administering step comprises subcutaneously injecting the antibody formulation.
28. A method for treating Psoriatic Arthritis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Psoriatic Arthritis.
29. The method of claim 28, wherein the subject is a human being.
30. The method of claim 28 or 29, wherein the administering step comprises subcutaneously injecting the antibody formulation.

31. A method for treating Ankylosing Spondylitis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Ankylosing Spondylitis.
32. The method of claim 31, wherein the subject is a human being.
33. The method of claim 31 or 32, wherein the administering step comprises subcutaneously injecting the antibody formulation.
34. A method for treating Crohn's Disease, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Crohn's Disease.
35. The method of claim 34, wherein the subject is a human being.
36. The method of claim 34 or 35, wherein the administering step comprises subcutaneously injecting the antibody formulation.
37. A method for treating Plaque Psoriasis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Plaque Psoriasis.
38. The method of claim 37, wherein the subject is a human being.
39. The method of claim 37 or 38, wherein the administering step comprises subcutaneously injecting the antibody formulation.
40. A method for treating Ulcerative Colitis, comprising administering to a subject in need thereof the buffered antibody formulation of any one of claims 1-21 in an amount effective to treat Ulcerative Colitis.
41. The method of claim 40, wherein the subject is a human being.

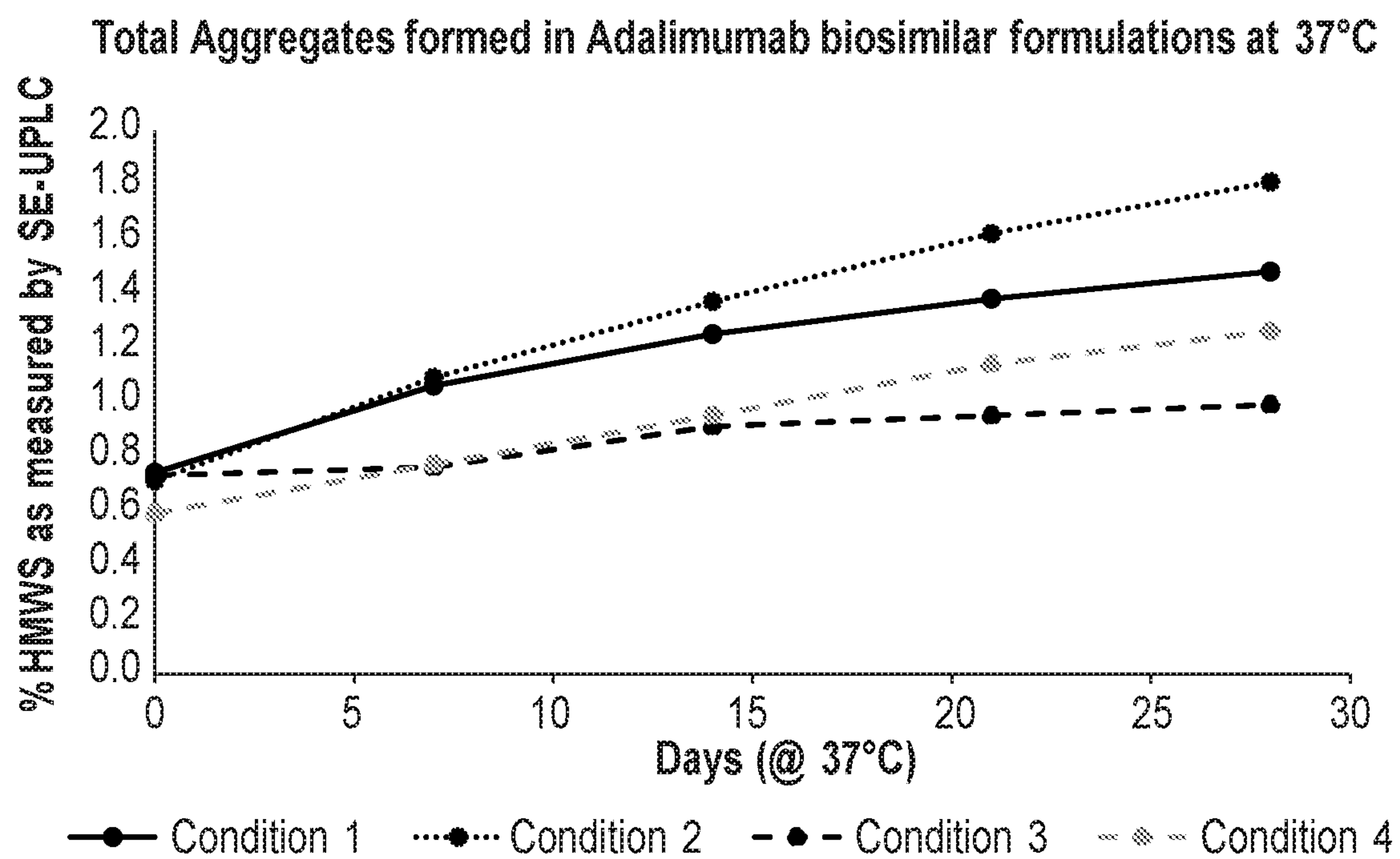
42. The method of claim 40 or 41, wherein the administering step comprises subcutaneously injecting the antibody formulation.
43. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Rheumatoid Arthritis.
44. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Juvenile Idiopathic Arthritis.
45. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Psoriatic Arthritis.
46. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Ankylosing Spondylitis.
47. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Crohn's Disease.
48. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Plaque Psoriasis.
49. The buffered antibody formulation of any one of claims 1-21 for use in the treatment of Ulcerative Colitis.
50. The buffered antibody formulation of any one of claims 1-21 for use in the manufacture of a medicament.
51. The buffered antibody formulation of any one of claims 1-21 for use in the manufacture of a medicament for the treatment of Rheumatoid Arthritis, Juvenile Idiopathic Arthritis, Psoriatic Arthritis, Ankylosing Spondylitis, Crohn's Disease, Plaque Psoriasis, or Ulcerative Colitis.

52. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Rheumatoid Arthritis.
53. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Juvenile Idiopathic Arthritis.
54. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Psoriatic Arthritis.
55. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Ankylosing Spondylitis.
56. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Crohn's Disease.
57. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Plaque Psoriasis.
58. A kit, comprising the buffered antibody formulation of any one of claims 1-21 and instructions for using the antibody formulation in a method for treating Ulcerative Colitis.
59. The kit of any one of claims 52-58, further comprising a device for injecting the antibody formulation into a subject.
60. The kit of claim 59, wherein the device comprises a syringe and a needle.
61. The kit of claim 59, wherein the device comprises a catheter.

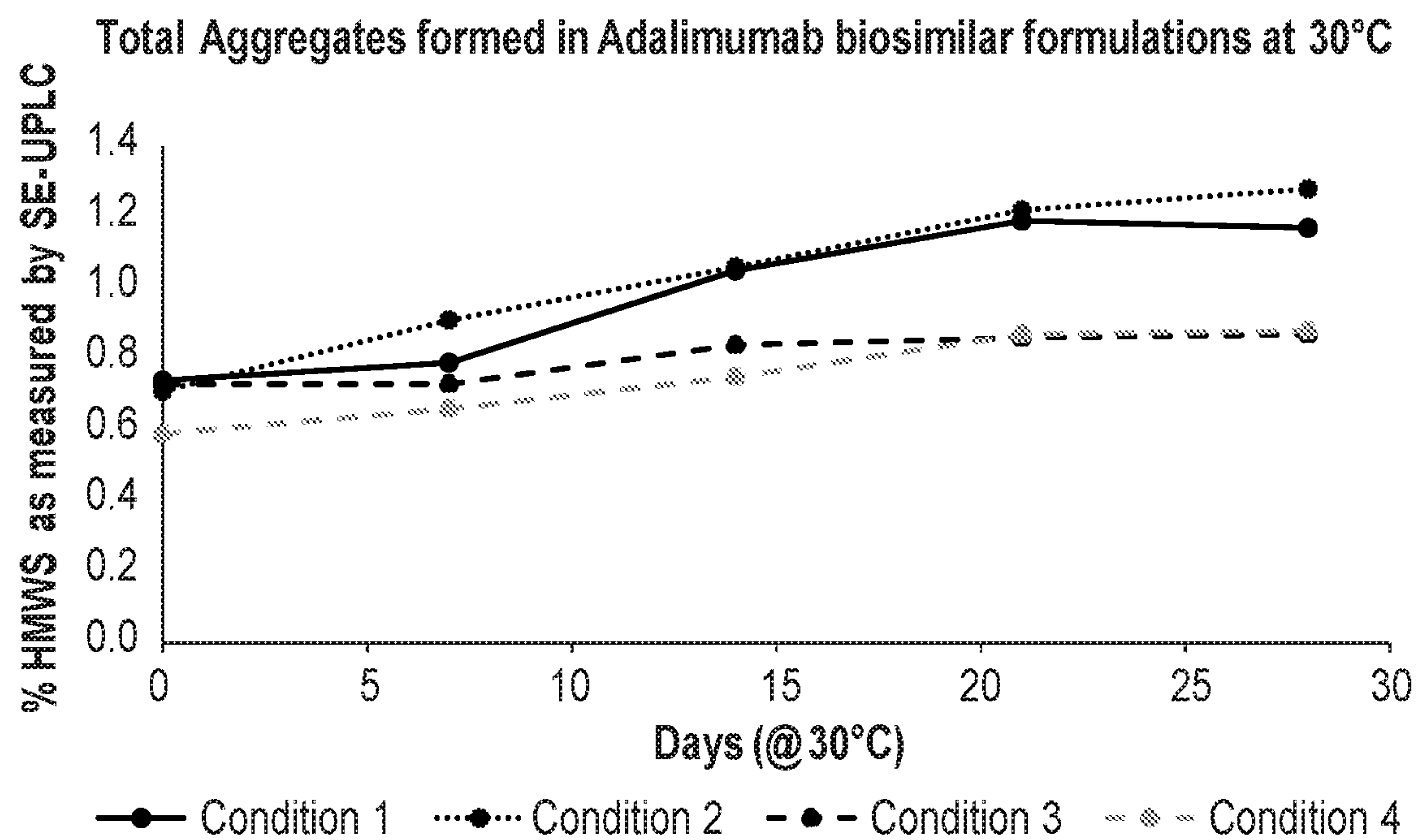
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FIGURE 1

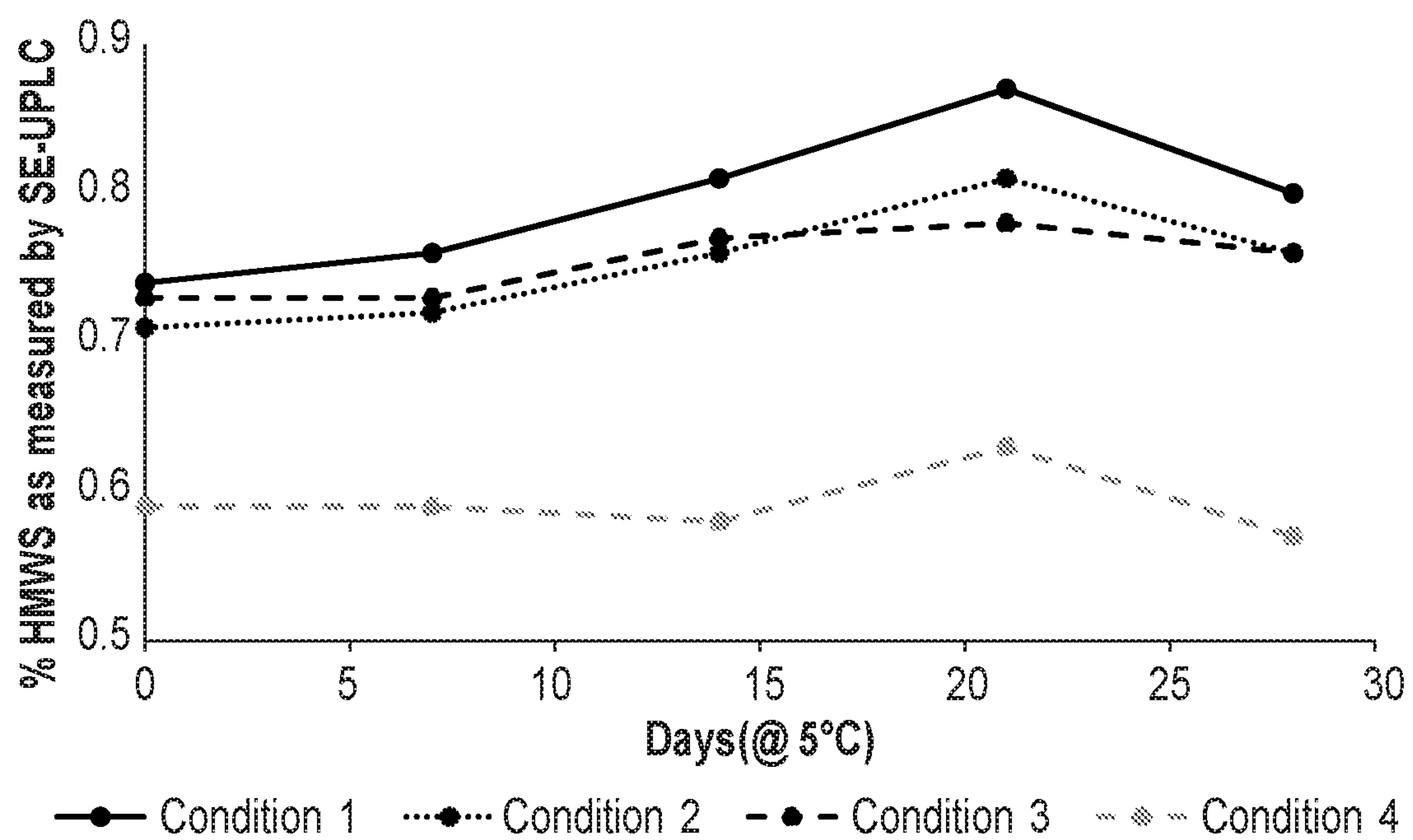
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FIGURE 2

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FIGURE 3

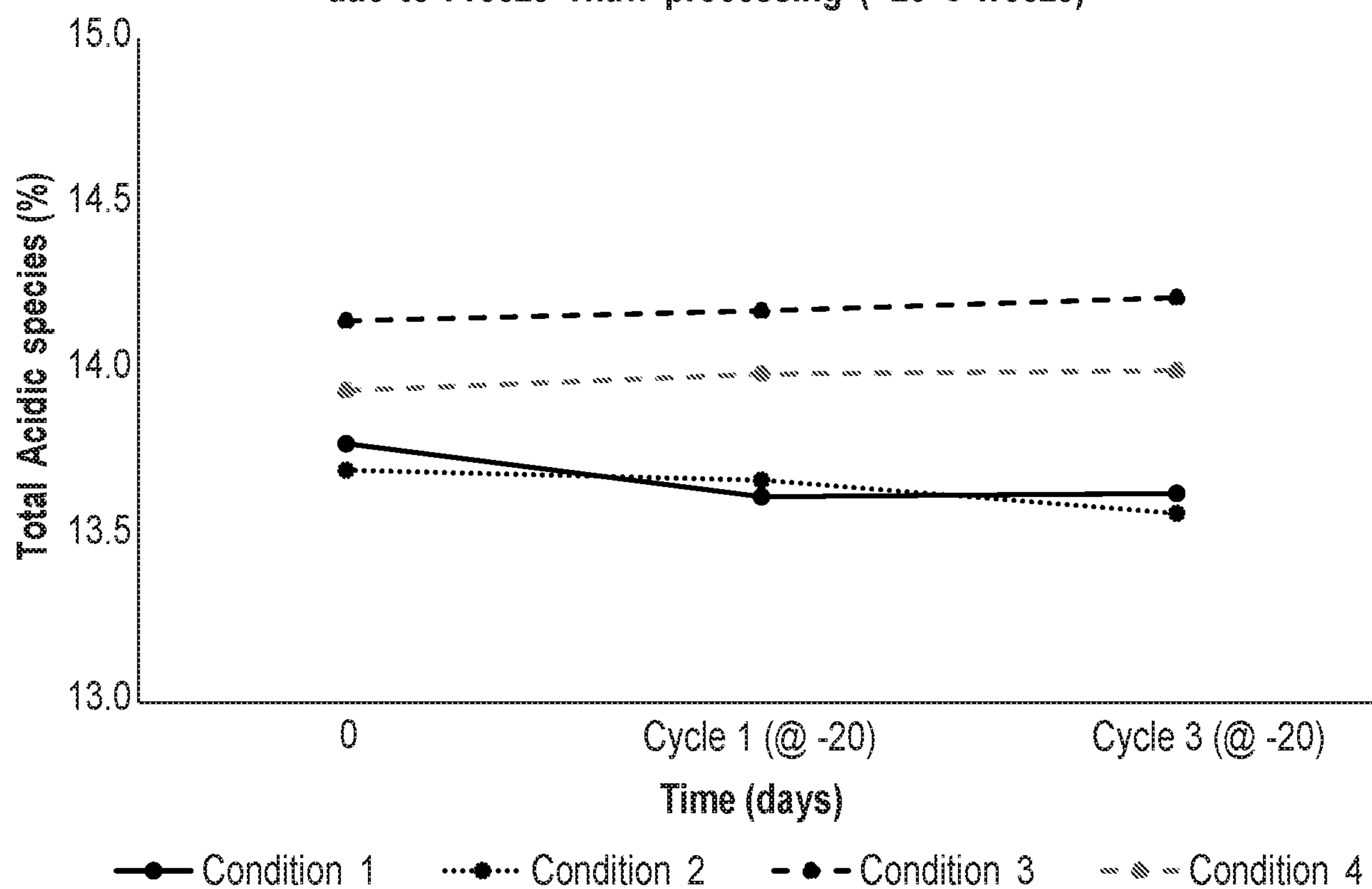
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FIGURE 4**Total Aggregates formed in Adalimumab biosimilar formulations at 5°C**

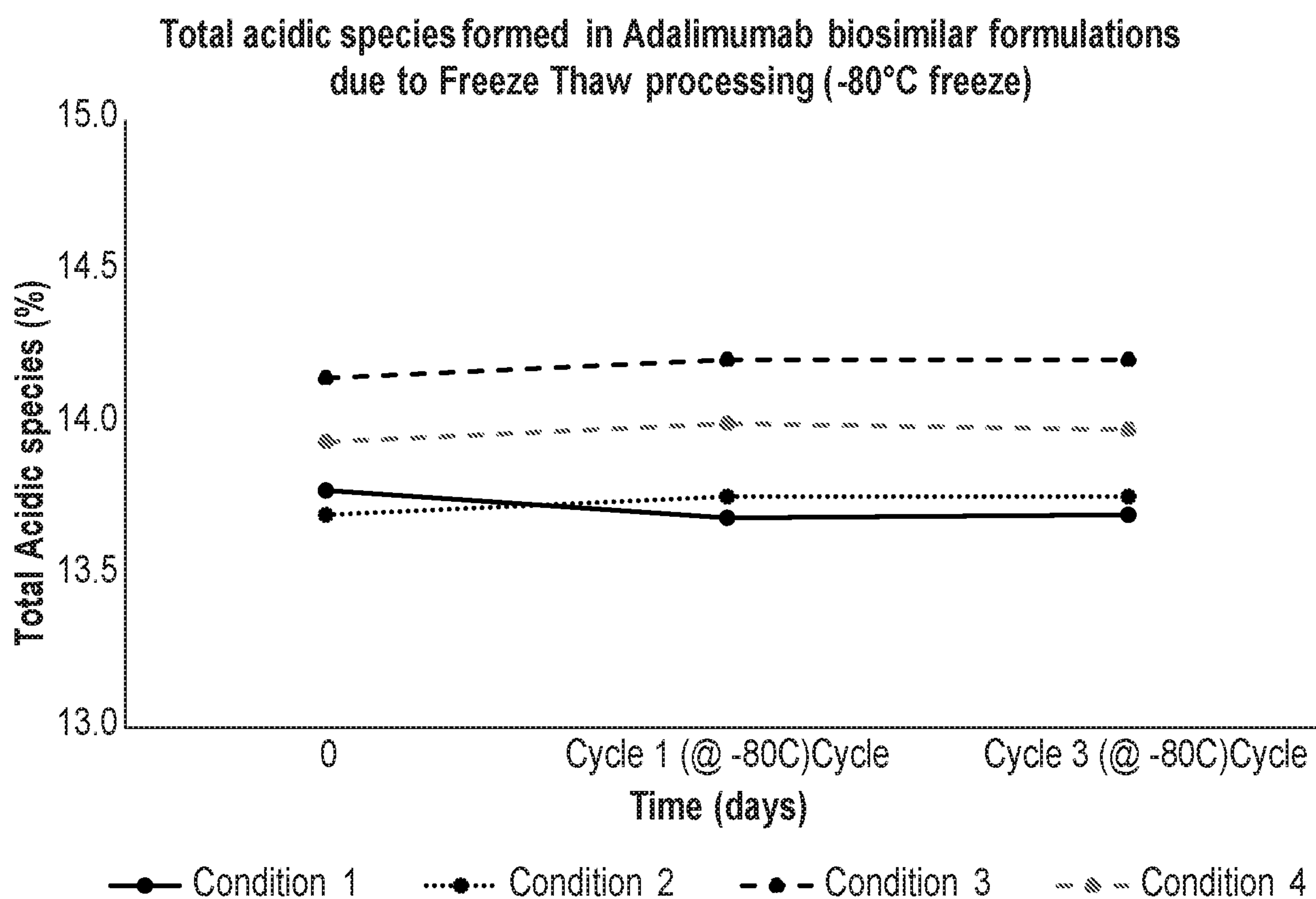
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FIGURE 5

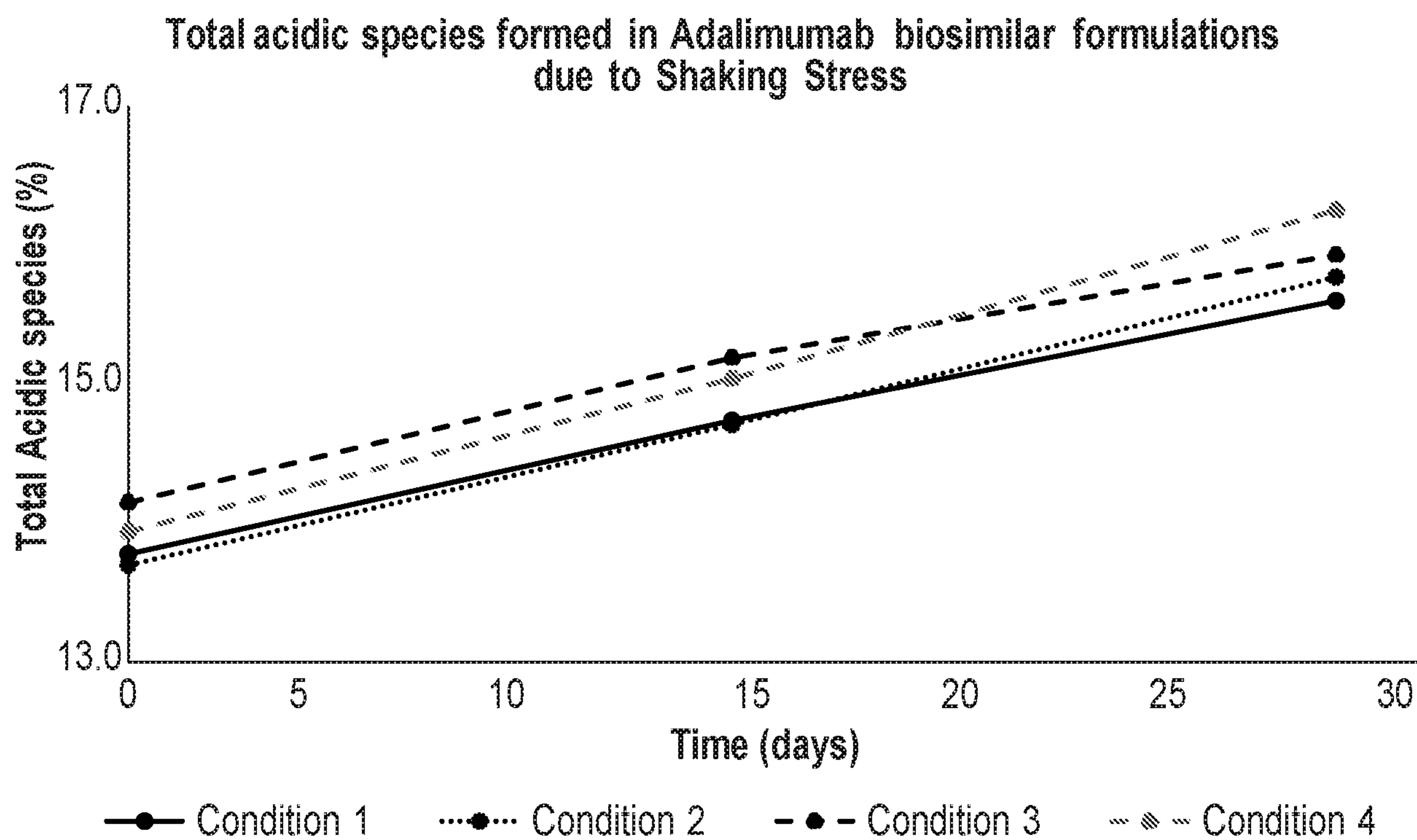
Total acidic species formed in Adalimumab biosimilar formulations
due to Freeze Thaw processing (-20°C freeze)



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FIGURE 6

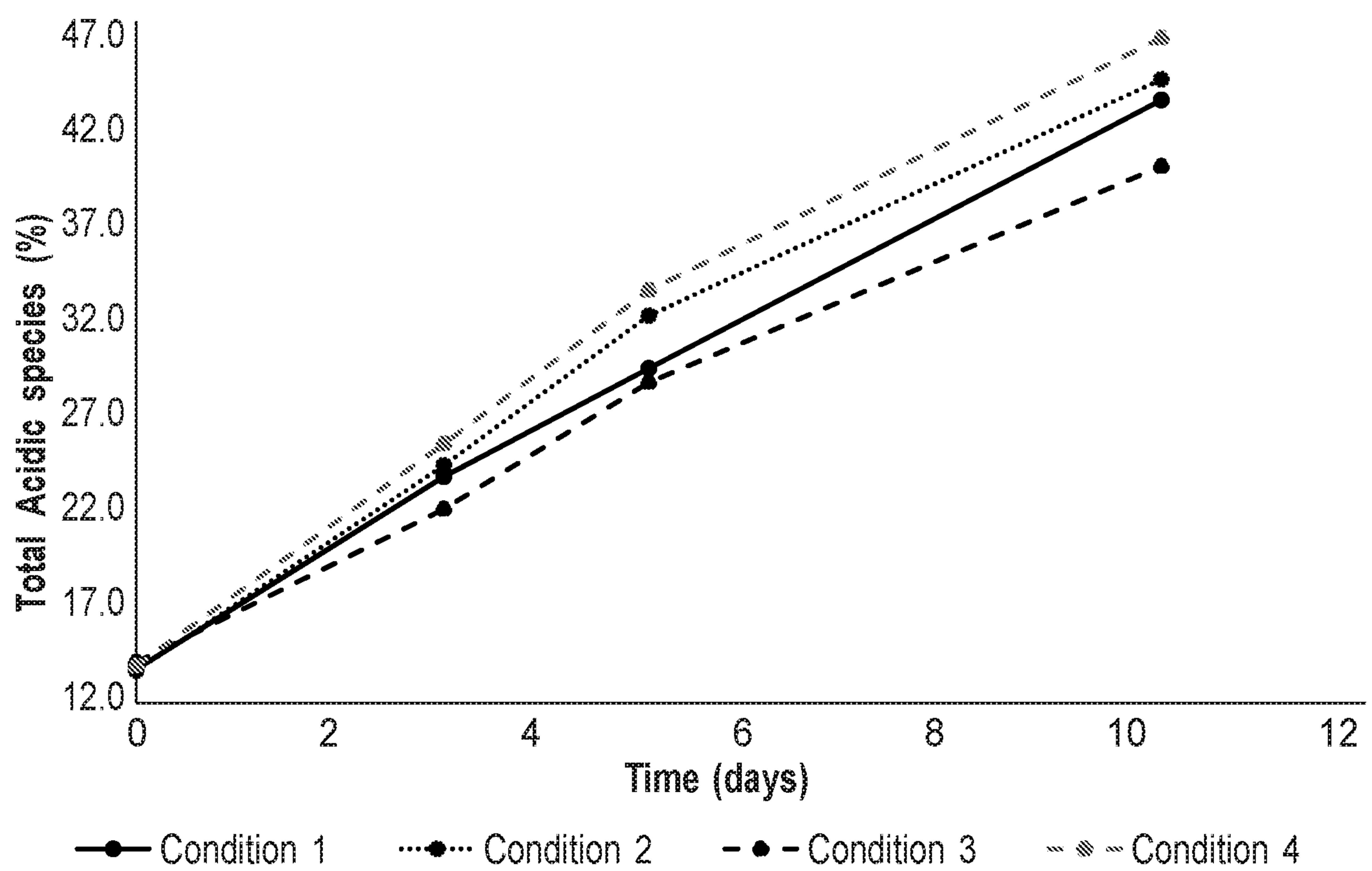
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FIGURE 7

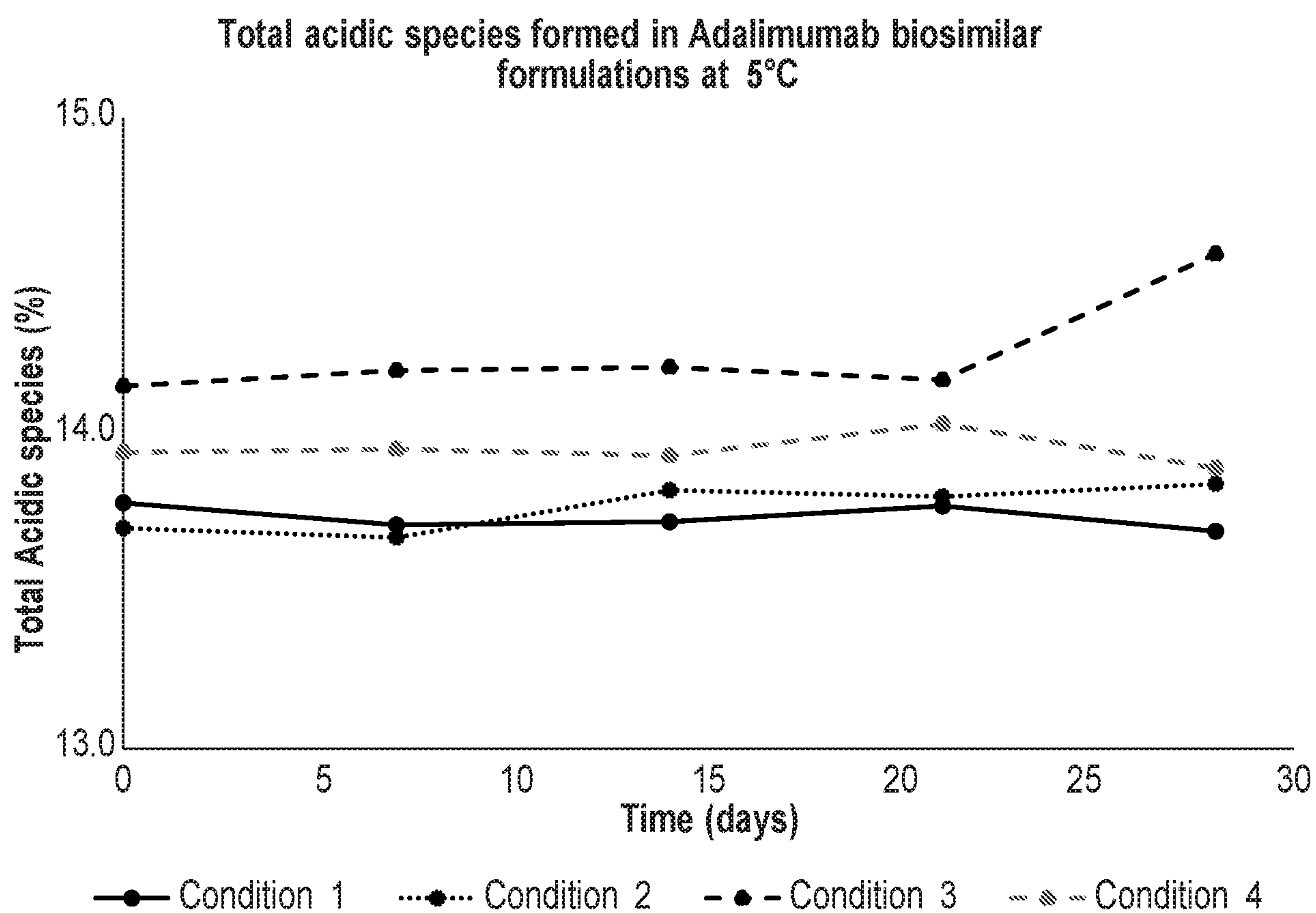
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FIGURE 8

Total acidic species formed in Adalimumab biosimilar formulations
at 55°C



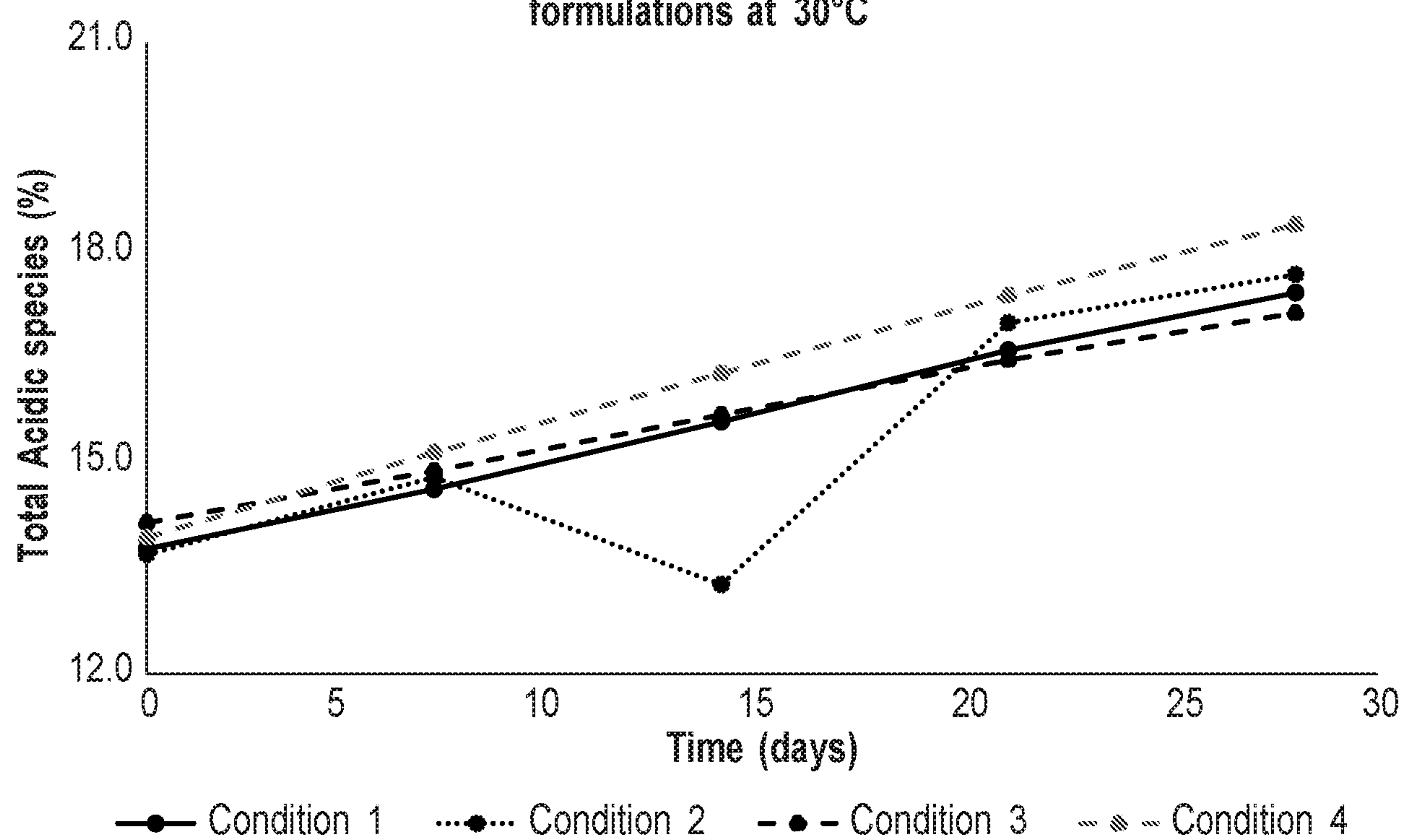
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FIGURE 9

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FIGURE 10

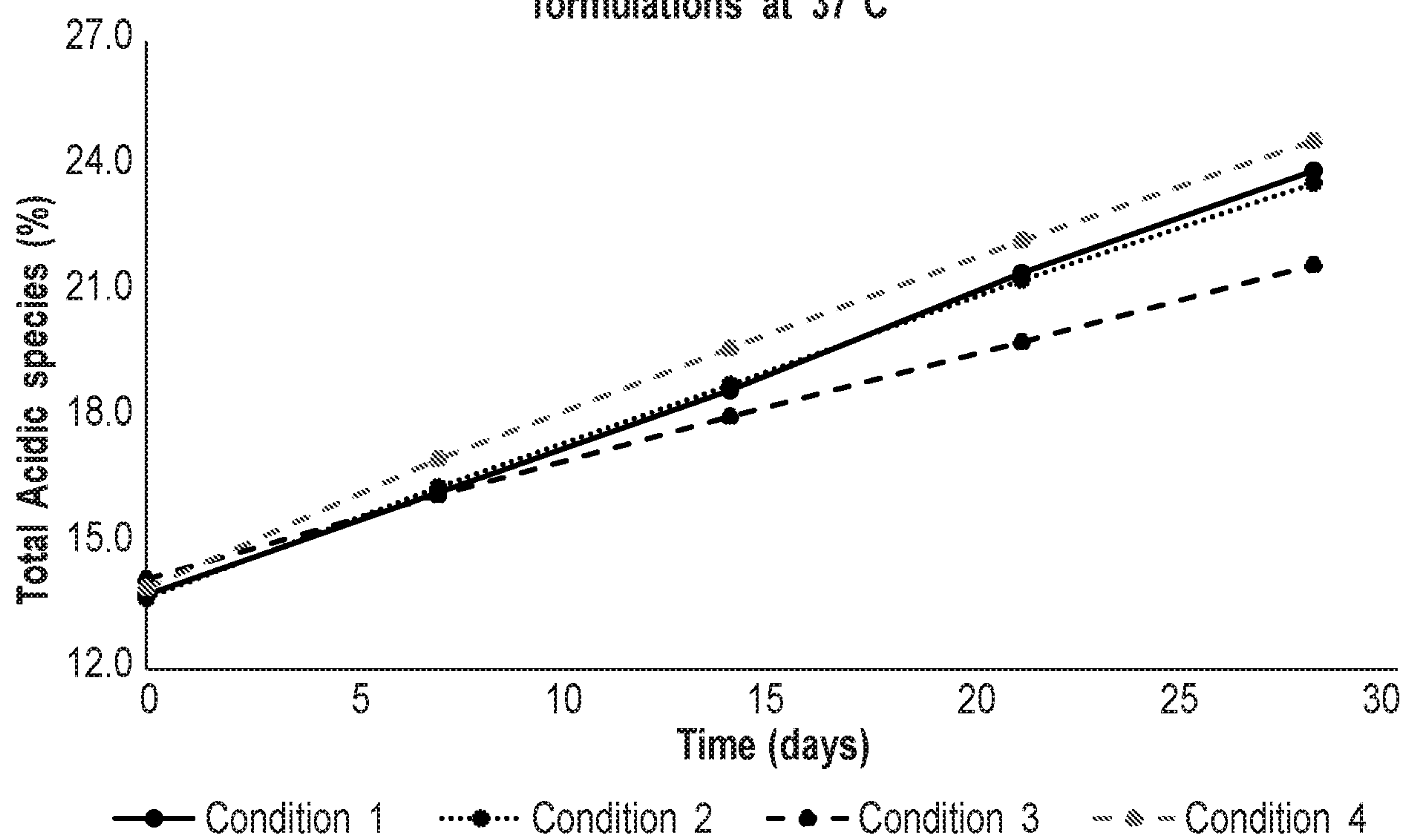
Total acidic species formed in Adalimumab biosimilar formulations at 30°C



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FIGURE 11

Total acidic species formed in Adalimumab biosimilar formulations at 37°C



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FIGURE 12

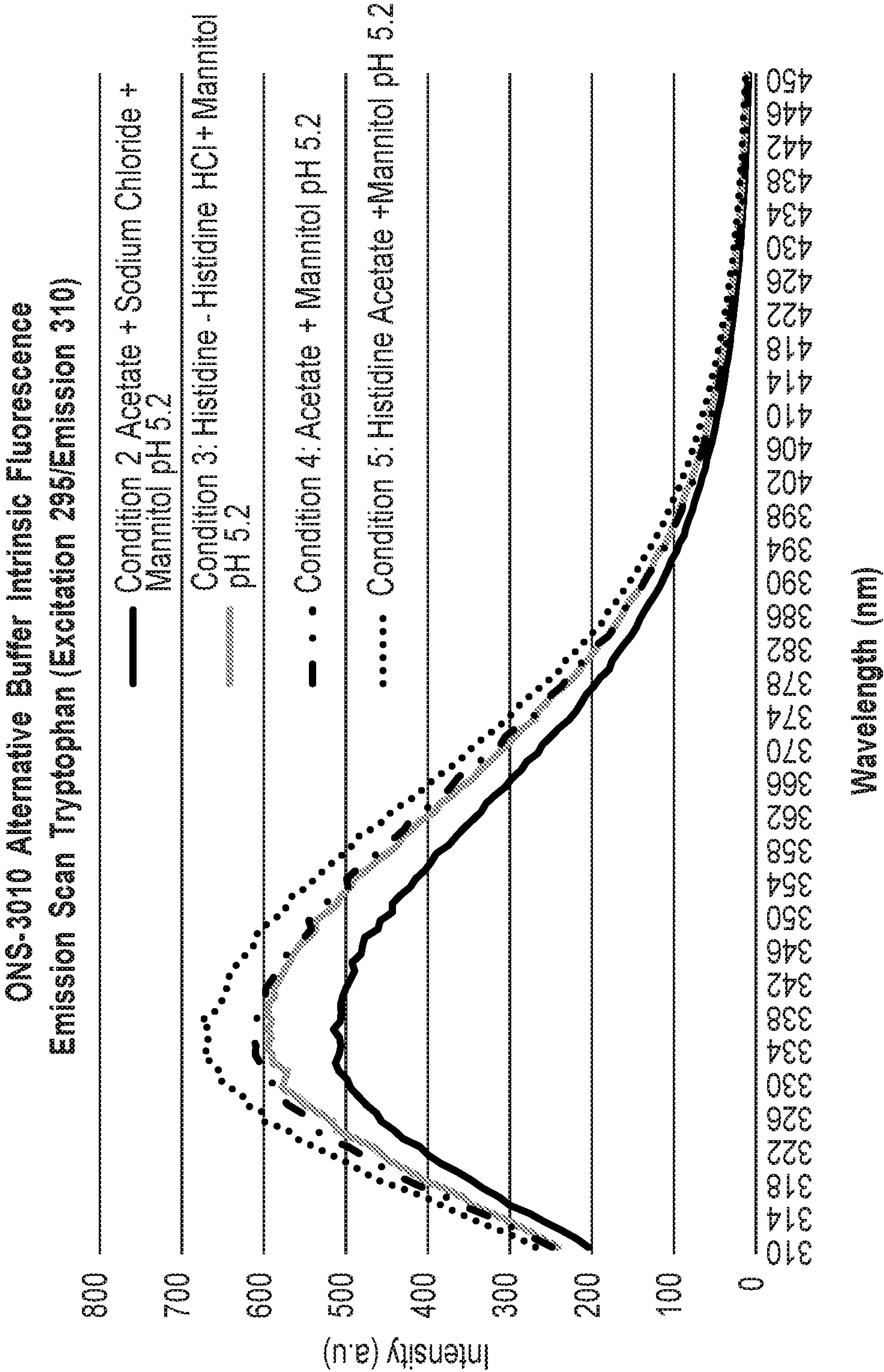


FIGURE 13
DLS RH VS CONCENTRATION

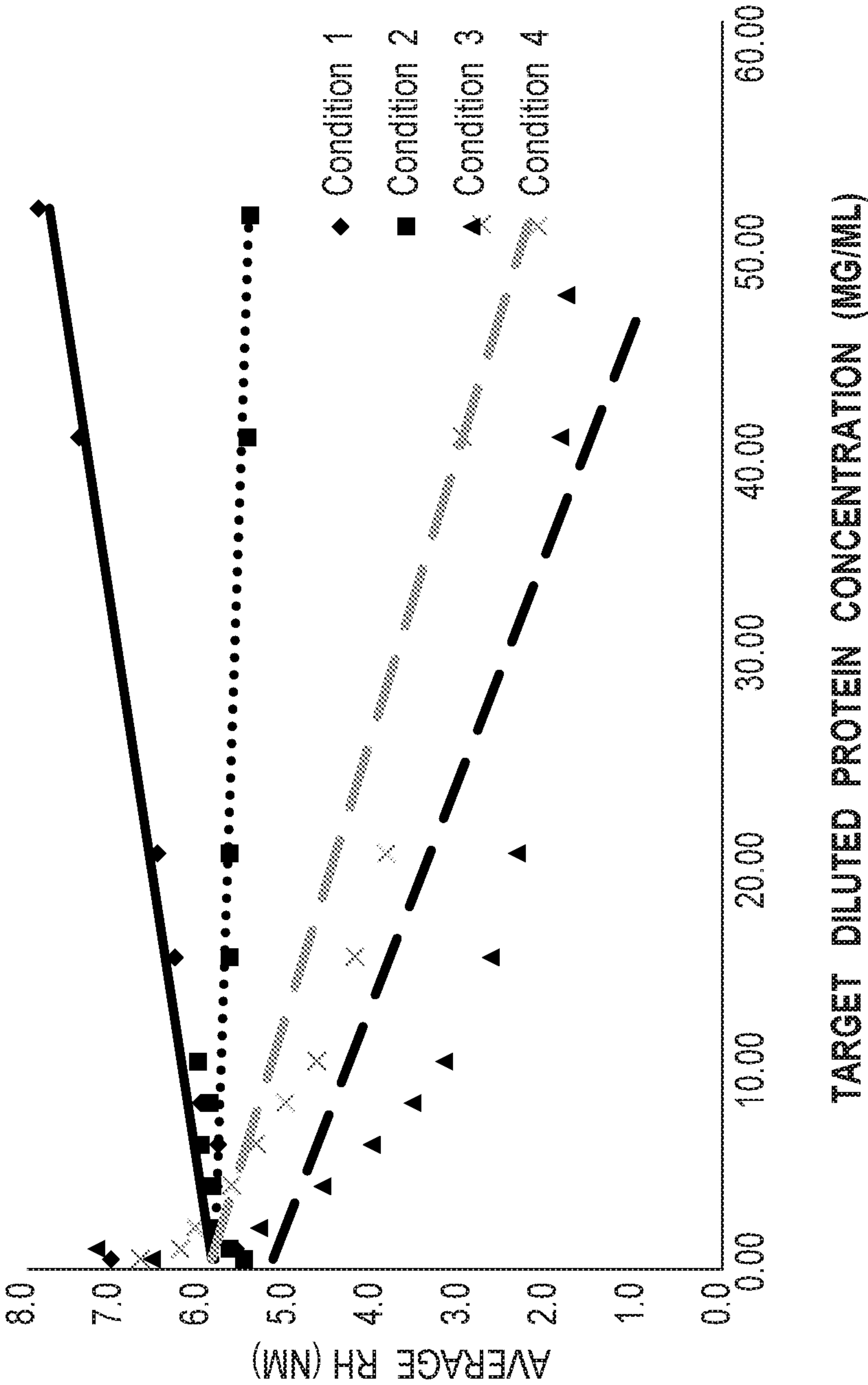
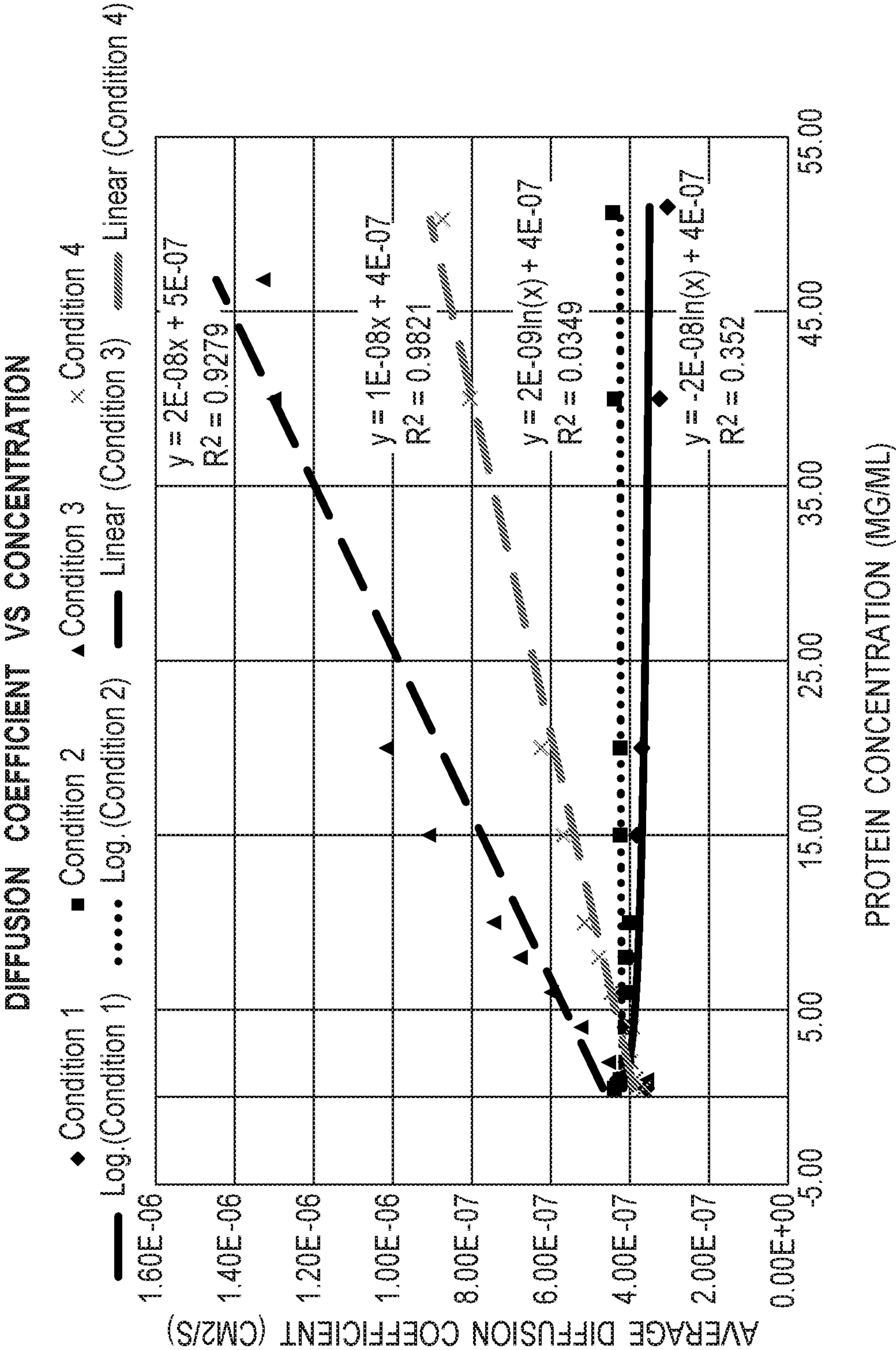


FIGURE 14



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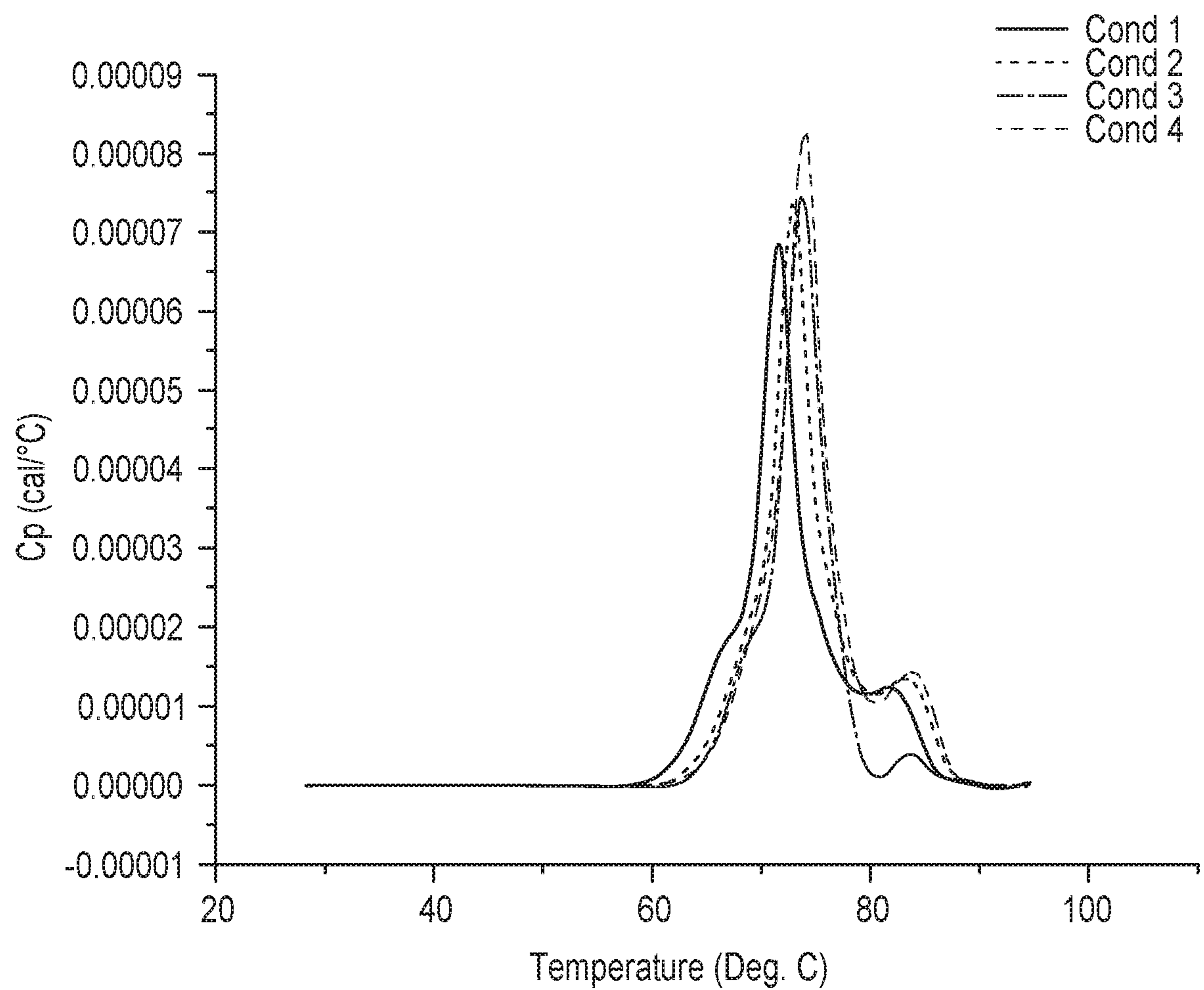
FIGURE 15

FIGURE 16

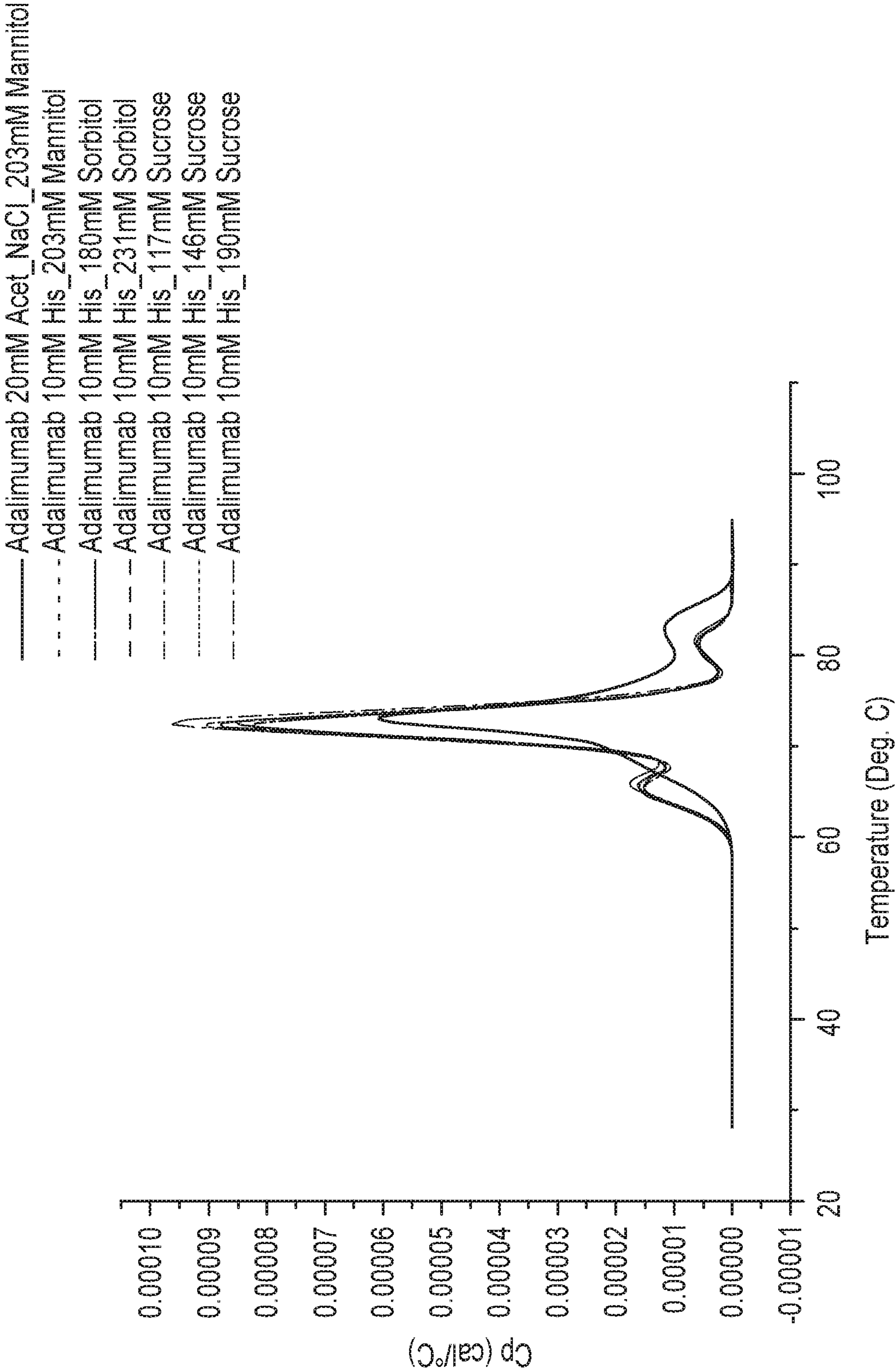


FIGURE 17

Colloidal stability of Adalimumab in Histidine
buffer-effect of buffer molarity

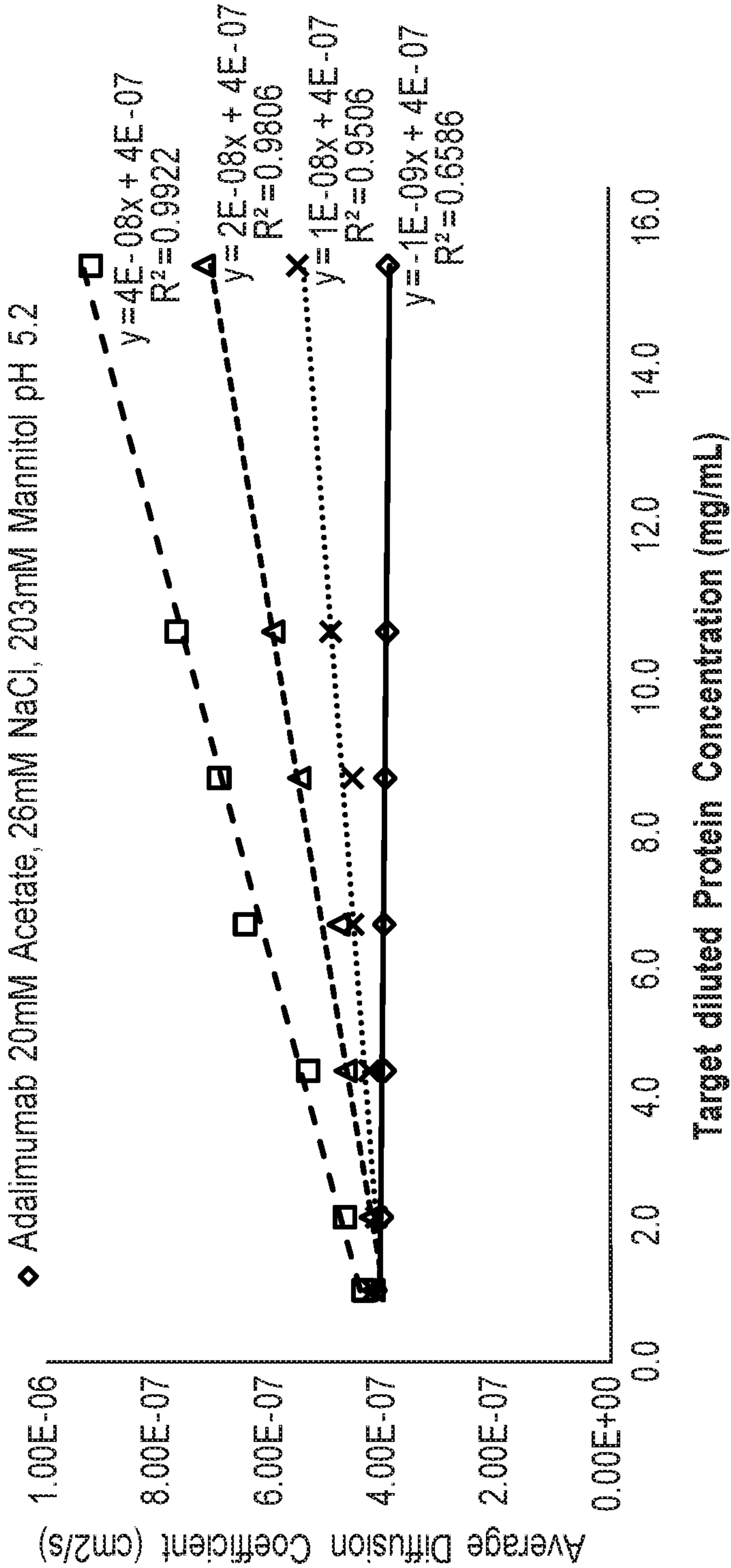


FIGURE 18

Colloidal stability of Adalimumab in Histidine buffer
-effect of buffer molarity

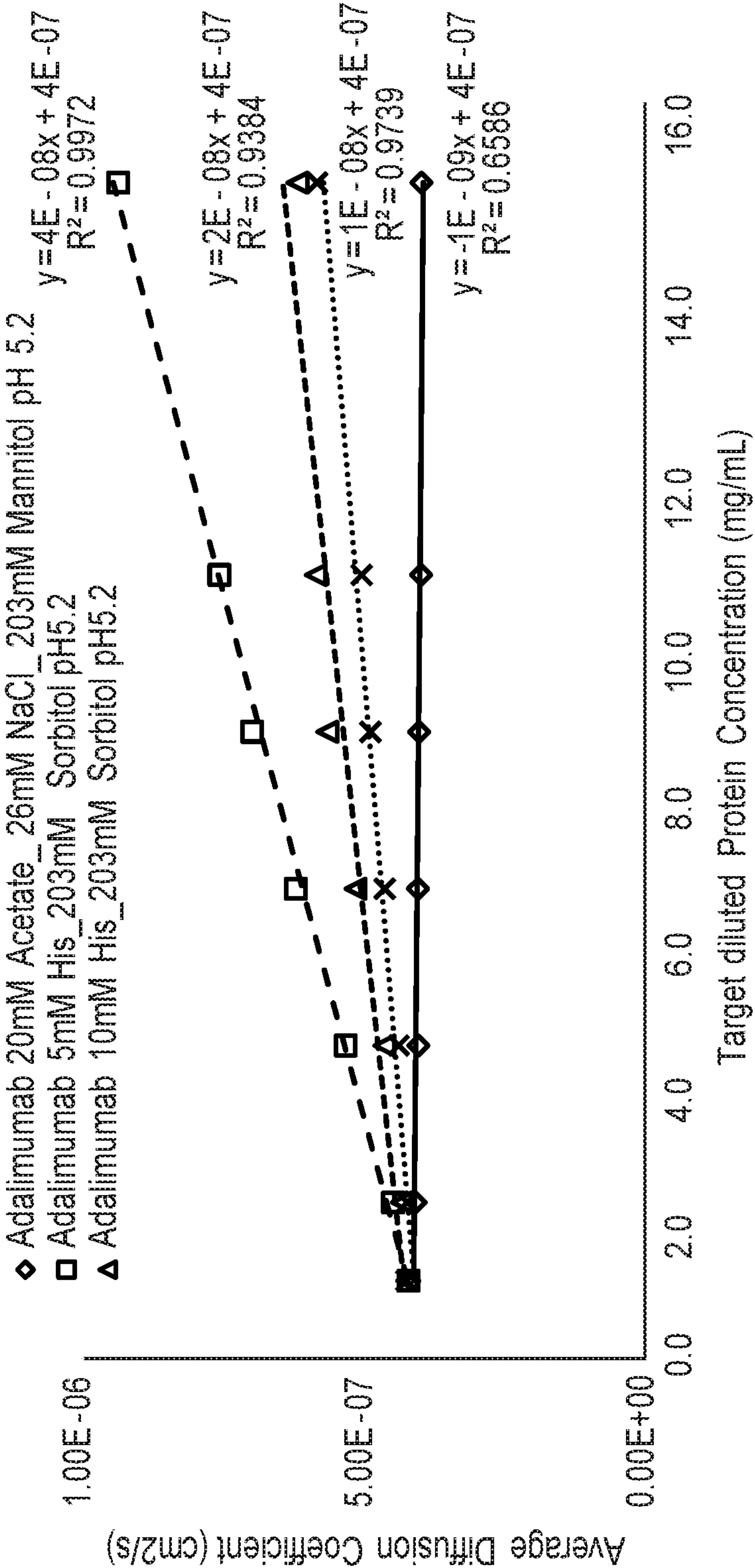
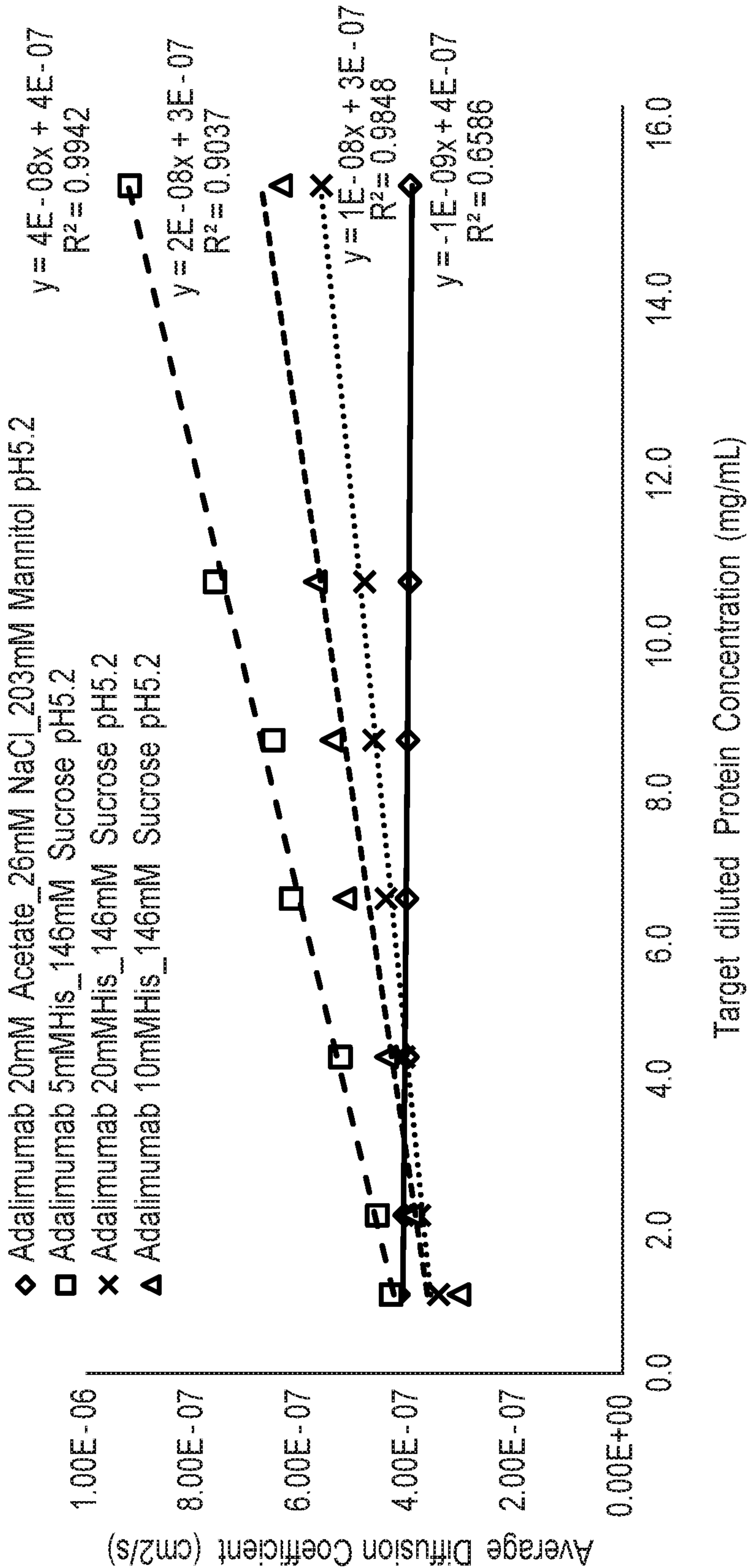


FIGURE 19

Colloidal stability of Adalimumab in Histidine buffer-
effect of buffer molarity



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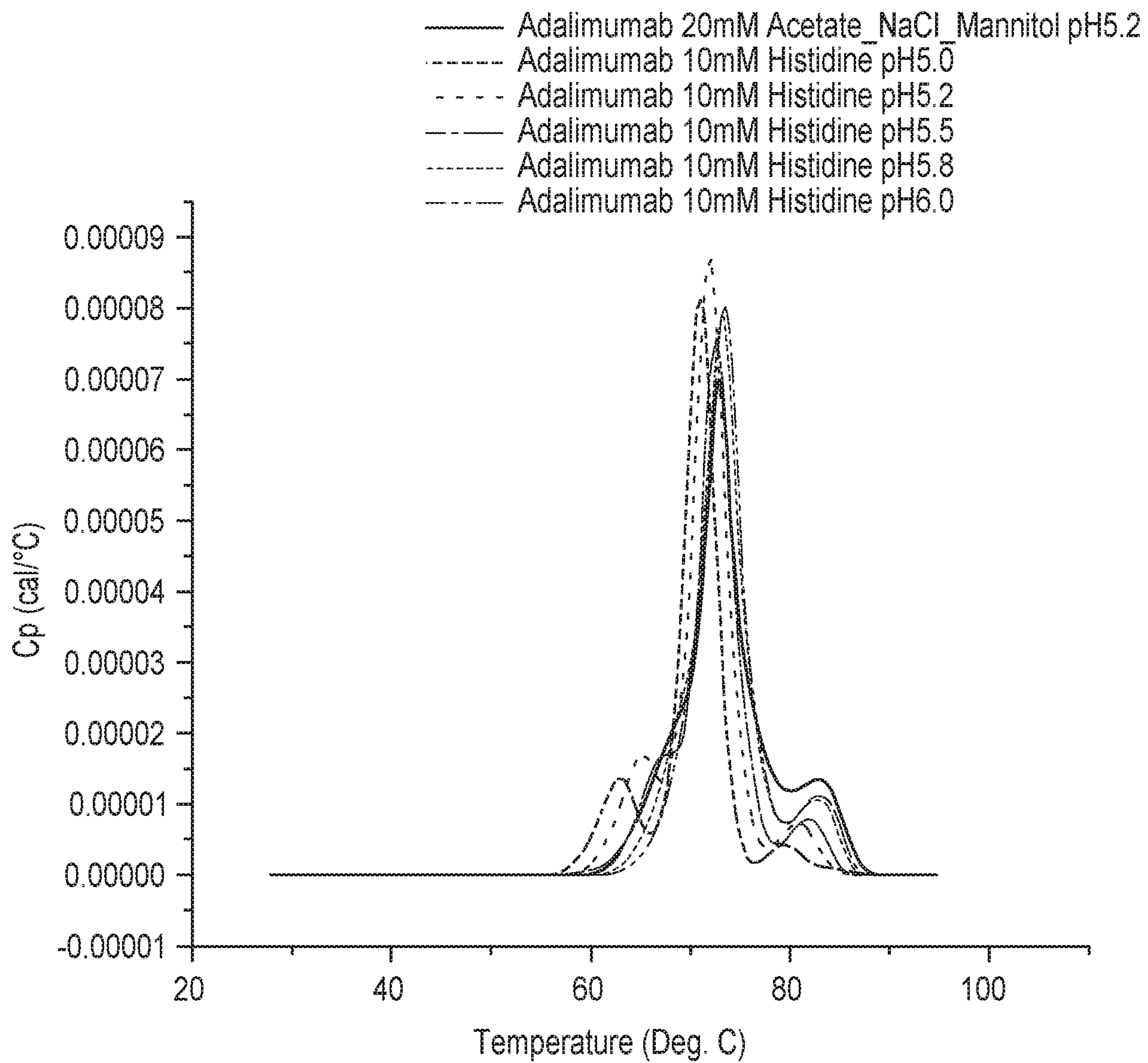
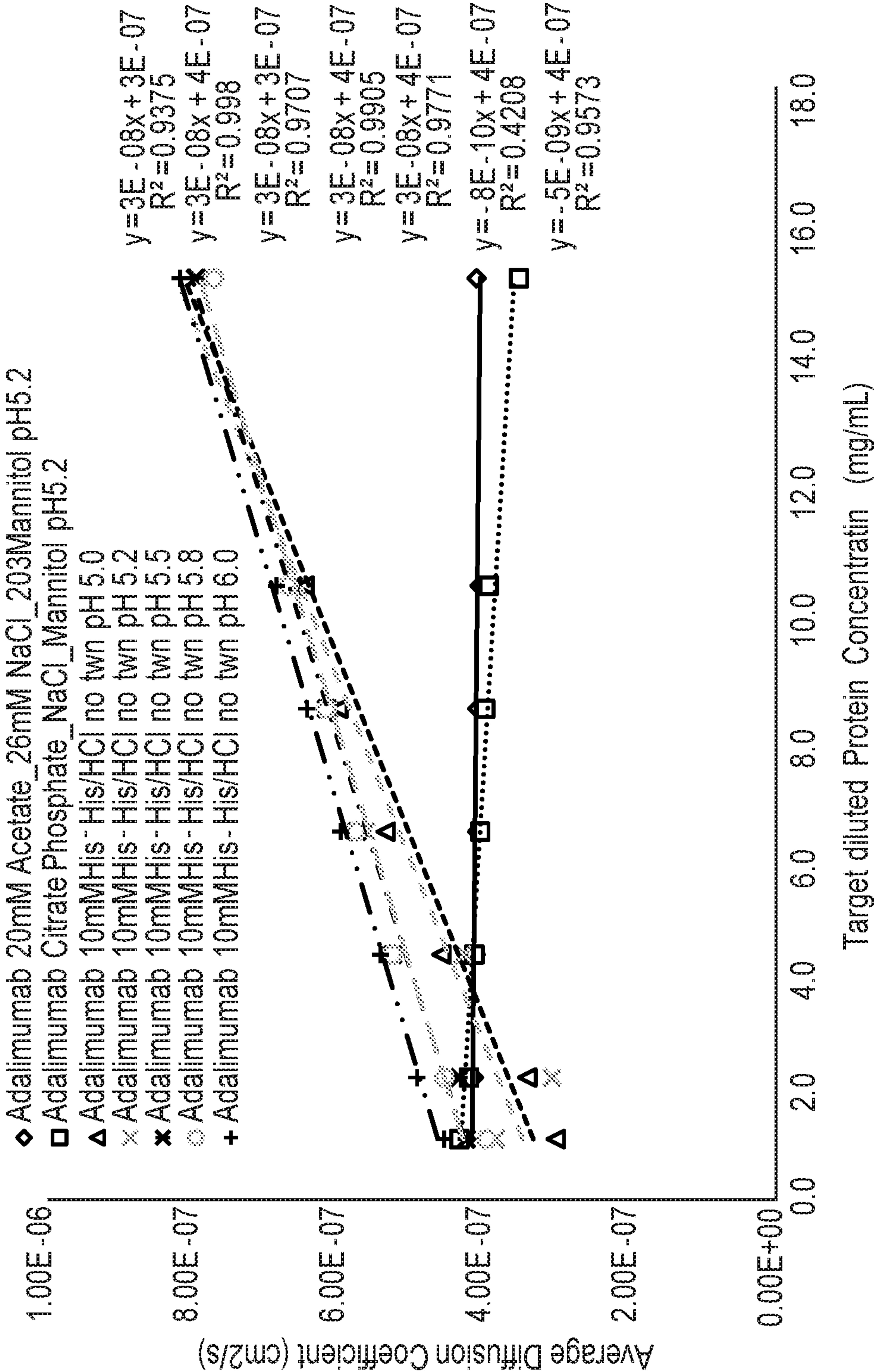
FIGURE 20

FIGURE 21

Colloidal stability of Adalimumab in Histidine
buffer pH range - 5 to 6



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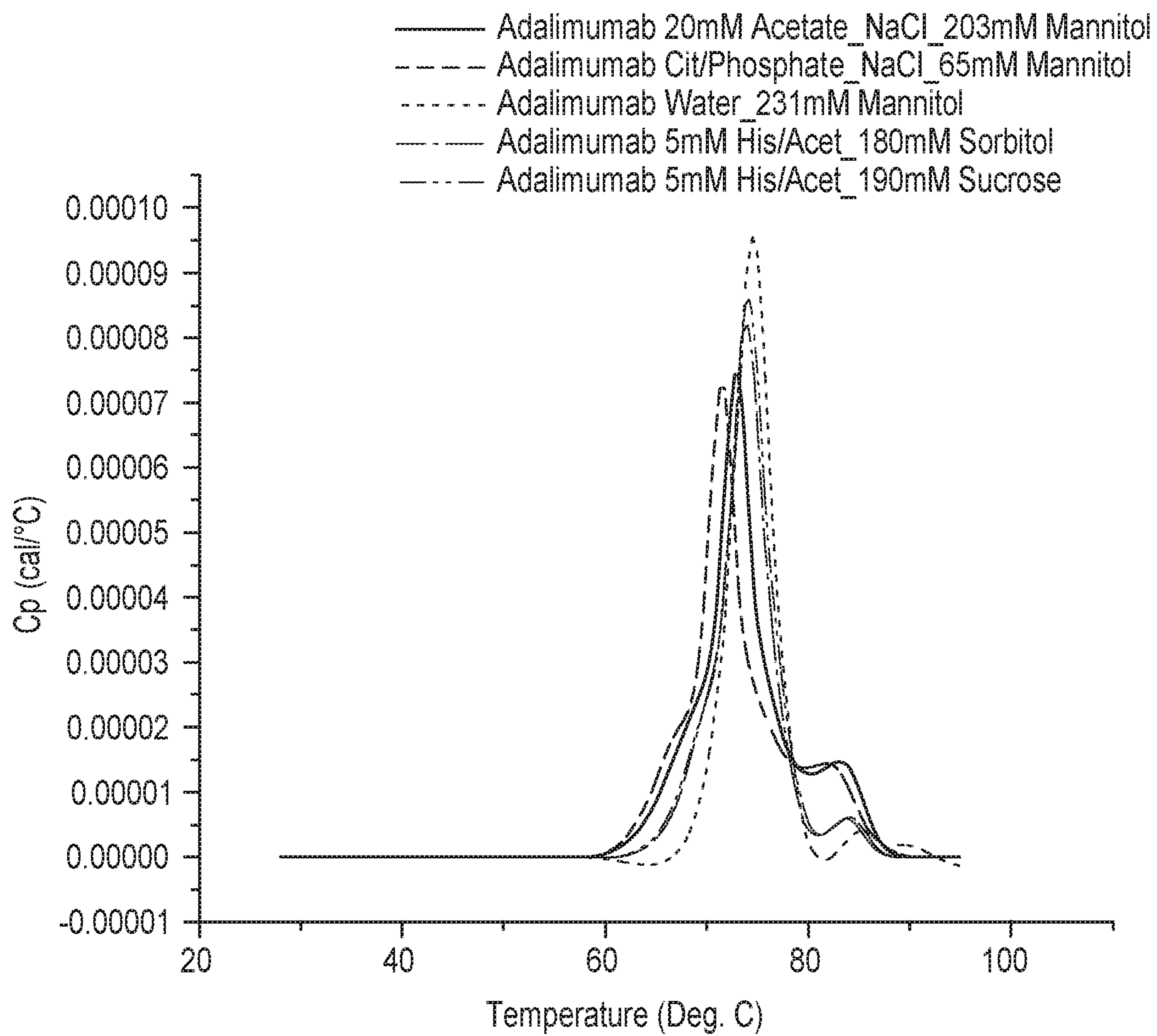
FIGURE 22

FIGURE 23
Colloidal stability of Adalimumab in Histidine -
Acetate buffer

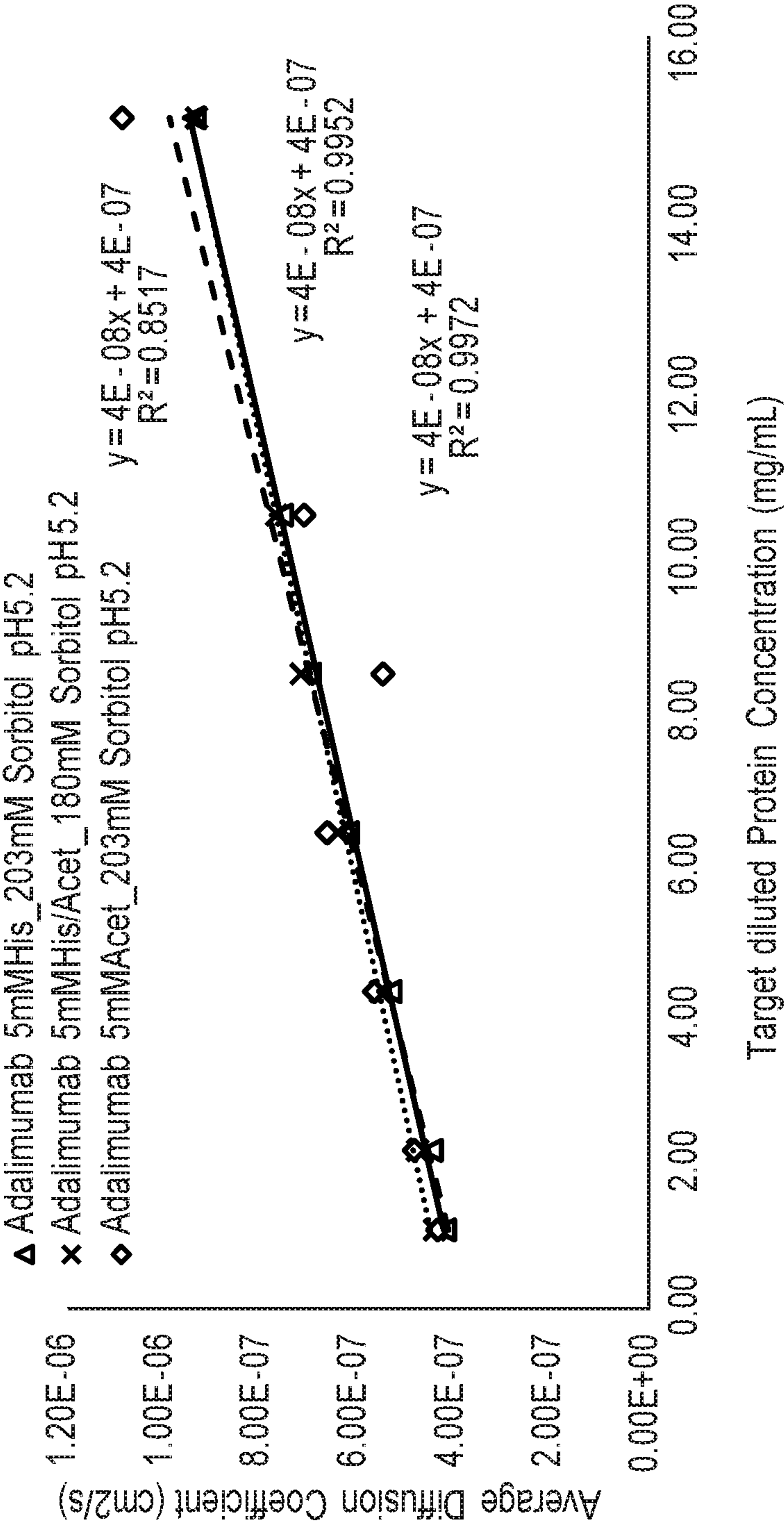


FIGURE 24

Colloidal Stability of Adalimumab in Histidine -
Acetate buffer

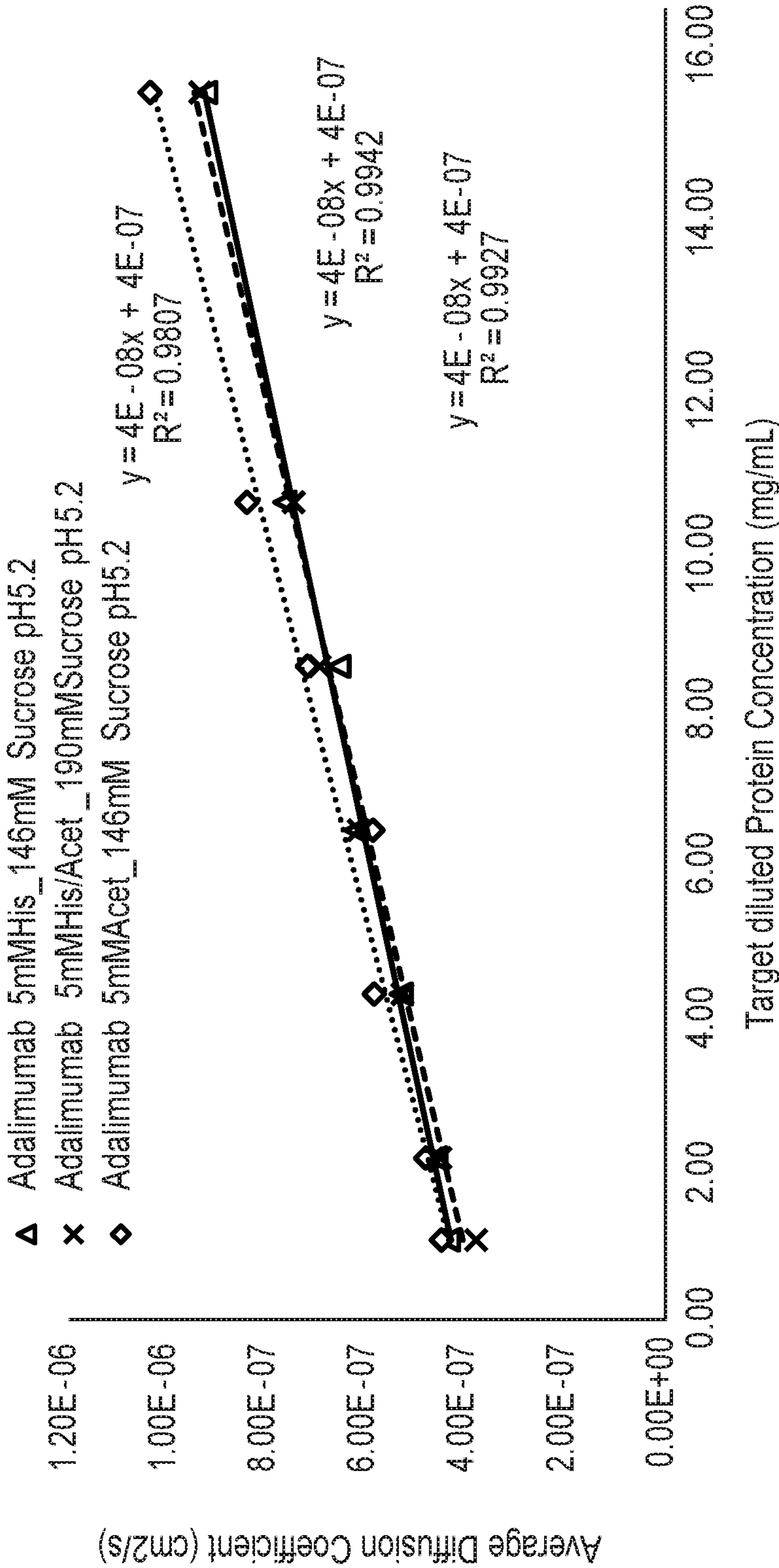


FIGURE 16

