

We claim: Claims:

- 1) A method of producing an anti-CD38 antibody expressed from a polynucleotide encoding a heavy chain variable region (VH) of SEQ ID NO: 7 and a light chain variable region (VL) of SEQ ID NO: 8 having an asialo, mono-galacto core-fucosylated biantennary glycan (G1F) oligosaccharide content between ~~about~~ 15% and ~~about~~ 27%, comprising:
 - a) preparing a culture medium comprising ~~about 8.5~~ between 2 parts per billion (ppb) ~~or less and 8.5 ppb~~ manganese (Mn); and
 - b) controlling the G1F oligosaccharide content of the anti-CD38 antibody by culturing a host cell comprising the polynucleotide encoding the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 in the culture medium prepared in step a), thereby producing the anti-CD38 antibody expressed from the polynucleotide encoding the -VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 having the G1F oligosaccharide content between ~~about~~ 15% and ~~about~~ 27%.
- 2) The A method of claim 1, wherein the G1F oligosaccharide content of the ~~producing a drug product~~ comprising:
 - a) producing an anti-CD38 antibody is between about 15% ~~according to the method of claim 1; and about 25%.~~
 - 3) The method of claim 1, wherein the G1F oligosaccharide content of b) formulating the anti-CD38 antibody is between about 21% and about 25% as a drug product.
- 4) ~~The method of any one of claims 1-3, wherein a G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%.~~
- 5) ~~The method of any one of claims 1-3, wherein the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and 74%.~~
- 3) The method of claim 2, wherein formulating the drug product comprises formulating the anti-CD38 antibody at from 20 mg/mL to 180 mg/mL with recombinant human hyaluronidase (rHuPH20) in an amount of from 30,000 U to 45,000 U, histidine at a concentration of from 5 mM to 15 mM, sorbitol at a concentration of from 100 mM to 300 mM, PS-20 at a concentration of from 0.01% w/v to 0.04 % w/v and methionine at a concentration of from 1 mg/mL to 2 mg/mL, at a pH of 5.0 to 6.0.
- 4) The method of claim 2 or claim 3, wherein formulating the drug product comprises formulating the anti-CD38 antibody at 120 mg/mL in 2,000 U/ml recombinant human hyaluronidase (rHuPH20), 5 mM to 15 mM histidine, 100 mM to 300 mM sorbitol, 0.01% w/v to 0.04 % w/v PS-20 and 1 mg/mL to 2 mg/mL methionine, at a pH of 5.6.

- 5) The method of any one of claims 2-4, wherein formulating the drug product comprises formulating the anti-CD38 antibody at 20 mg/ml in 25 mM acetic acid, 60 mM sodium chloride, 140 mM mannitol and 0.04% w/v polysorbate-20 (PS-20); at pH 5.5.
- 6) The method of any one of claims 1-5, wherein preparing the culture medium comprises measuring Mn concentration in one or more batches of raw material components used to prepare the culture medium and selecting those one or more batches of raw material components that in combination contain ~~about~~ between 2 ppb and 8.5 ppb or less Mn.
- 7) The method of claim 6, wherein the culture medium is prepared to comprise ~~between about 4.0 ppb and about 8.5 ppb Mn;~~ between 4.0 ppb and 6.5 ppb Mn; between 5.0 ppb and 6.5 ppb Mn; or between 2 ppb and 6.5 ppb Mn.
- 8) ~~The~~ A method of claim ~~producing an anti-CD38 antibody expressed from a polynucleotide encoding the VH of SEQ ID NO: 7, wherein the~~ producing an anti-CD38 antibody expressed from a polynucleotide encoding the VH of SEQ ID NO: 7, wherein the ~~and a VL of SEQ ID NO: 8 having a G1F oligosaccharide content between 15% and 27%, comprising:~~
- a) culturing a host cell expressing the polynucleotide encoding the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 in conditions that the anti-CD38 antibody is produced; and
- b) controlling the G1F oligosaccharide content of the anti-CD38 antibody by monitoring the concentration of Mn in the culture medium is prepared during biosynthesis of the anti-CD38 antibody and regulating the concentration of Mn in the culture medium during biosynthesis of the anti-CD38 antibody, wherein the concentration of Mn in the culture medium is regulated to comprise between 2 ppb and 8.5 ppb Mn, thereby producing the anti-CD38 antibody having the G1F oligosaccharide content between 15% and 27%.
- 9) The method of claim 8, wherein the culture medium is regulated to comprise between about 4.0 ppb and about 8.5 ppb Mn; between 4.0 ppb and 6.5 ppb Mn;
~~The method of claim 8, wherein the culture medium is prepared to comprise;~~ between about 5.0 ppb and about 6.5 ppb Mn; or between 2 ppb and 6.5 ppb Mn.
- 10) The method of any one of claims 1-9, wherein the G1F oligosaccharide content of the anti-CD38 antibody is:
- a) between 15% and 25%; or
- b) between 21% and 25%.
- 11) The method of any one of claims 1-10, wherein an asialo, agalacto core-fucosylated biantennary glycan (G0F) oligosaccharide content of the anti-CD38 antibody is:
- a) between 65% and 74%; or
- b) between 68% and 74%.

~~12)~~ The method of any one of claims 1-11, wherein:

a) the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 27%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%, and the culture medium is prepared or regulated to comprise between about 4.0 ppb and about 8.5 ppb Mn;

~~11)~~ The method of any one of claims 1-9, wherein b) the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is prepared or regulated to comprise between about 4.0 ppb and about 6.5 ppb Mn; or

~~12)~~ The method of any one of claims 1-9, wherein c) the G1F oligosaccharide content of the anti-CD38 antibody is between about 21% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is prepared or regulated to comprise between about 5.0 ppb and about 6.5 ppb Mn.

13) The method of any one of claims 1-12, wherein:

a) the culture medium is a basal medium or a feed medium; and/or

~~14)~~ The method of any one of claims 1-13, wherein b) culturing comprises a fed-batch culture or a perfusion culture; and/or

~~15)~~ The method of any one of claims 1-14, wherein the c) the host cell is an eukaryotic cell;

~~16)~~ The method of claim 16, optionally wherein the eukaryotic cell is a CHO cell, a PER.C6 cell, a NS0 cell, a Sp2/0 cell or a BHK cell. —

~~17)~~ The method of claim 16; optionally wherein the CHO cell is a CHO-K1 cell, a CHO-DG44 cell, a CHO-S cell or a CHO-DXB11 cell.

~~18)~~ The method of claim 17; and optionally further wherein the CHO cell is deficient in glutamine synthetase (GS).

~~19)~~ 14) The method of any one of claims 1-18, wherein the method is conducted under good manufacturing practice (GMP)-compliant conditions.

~~15)~~ The method of any one of claims 1-19, wherein the anti-CD38 antibody:

~~20)~~ a) comprises the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 9-8; and/or

~~21)~~ The method of any one of claims 1-20, wherein the anti-CD38 antibody b) comprises an IgG1 isotype; and/or

~~22)~~ The method of any one of claims 1-21, wherein the anti-CD38 antibody c) comprises a heavy chain (HC) of SEQ ID NO: 9 and a light chain (LC) of SEQ ID NO: 10; and/or

~~23)~~ The method of any one of claims 1-22, wherein the anti-CD38 antibody d) is a biosimilar; and/or e) is daratumumab.

- 24) A method of producing an anti-CD38 antibody expressed from a polynucleotide encoding the VH of SEQ ID NO: 7 and a VL of SEQ ID NO: 8 having a G1F oligosaccharide content between about 15% and about 27%, comprising:
- a) culturing a host cell expressing the polynucleotide encoding the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 in conditions that the anti-CD38 antibody is produced; and
 - b) controlling the G1F oligosaccharide content of the anti-CD38 antibody by monitoring the concentration of Mn in the culture medium during biosynthesis of the anti-CD38 antibody and regulating the concentration of Mn in the culture medium during biosynthesis of the anti-CD38 antibody, wherein the concentration of Mn in the culture medium is regulated to comprise about 8.5 ppb or less Mn, thereby producing the anti-CD38 antibody having the G1F oligosaccharide content between about 15% and about 27%.
- 25) The method of claim 24, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 25%.
- 26) The method of claim 25, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 21% and about 25%.
- 27) The method of claim 24, wherein a G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%.
- 28) The method of claim 25, wherein the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and 74%.
- 29) The method of claim 24, wherein the concentration of Mn in the culture medium is regulated to comprise between about 4.0 ppb and about 8.5 ppb Mn.
- 30) The method of claim 29, wherein the concentration of Mn in the culture medium is regulated to comprise between about 4.0 ppb and about 6.5 ppb Mn.
- 31) The method of claim 30, wherein the concentration of Mn in the culture medium is regulated to comprise between about 5.0 ppb and about 6.5 ppb Mn.
- 32) The method of any one of claims 24-30, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 27%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%, and the culture medium is regulated to comprise between about 4.0 ppb and about 8.5 ppb Mn.
- 33) The method of any one of claims 24-30, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is regulated to comprise between about 4.0 ppb and about 6.5 ppb Mn.

- 34) The method of any one of claims 24-30, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 21% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is regulated to comprise between about 5.0 ppb and about 6.5 ppb Mn.
- 35) The method of any one of claims 24-34, wherein the culture medium is a basal medium or a feed medium.
- 36) The method of any one of claims 24-35, wherein culturing comprises a fed batch culture or a perfusion culture.
- 37) The method of any one of claims 24-36, wherein the host cell is an eukaryotic cell.
- 38) The method of claim 37, wherein the eukaryotic cell is a CHO cell, a PER.C6 cell, a NS0 cell, a Sp2/0 cell or a BHK cell.
- 39) The method of claim 38, wherein the CHO cell is a CHO-K1 cell, a CHO-DG44 cell, a CHO-S cell or a CHO-DXB11 cell.
- 40) The method of claim 39, wherein the CHO cell is deficient in GS.
- 41) The method of any one of claims 24-40, wherein the method is conducted under GMP-compliant conditions.
- 42) The method of any one of claims 24-41, wherein the anti-CD38 antibody comprises the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 9.
- 43) The method of any one of claims 24-42, wherein the anti-CD38 antibody comprises an IgG1 isotype.
- 44) The method of any one of claims 24-43, wherein the anti-CD38 antibody comprises the HC of SEQ ID NO: 9 and the LC of SEQ ID NO: 10.
- 45) The method of any one of claims 24-44, wherein the anti-CD38 antibody is a biosimilar.
- 46) A method of producing a drug product comprising an anti-CD38 antibody expressed from a polynucleotide encoding the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 and having a G1F oligosaccharide content between about 15% and about 27%, comprising:
- a) preparing a culture medium comprising about 8.5 ppb or less Mn;
 - b) controlling the G1F oligosaccharide content of the anti-CD38 antibody by culturing a host cell comprising the polynucleotide encoding the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 8 in the culture medium prepared in step a), thereby producing the anti-CD38 antibody having the G1F oligosaccharide content between about 15% and about 27%; and
 - c) formulating the anti-CD38 antibody as a drug product.
- 47) The method of claim 46, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 25%.

- 48) The method of claim 47, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 21% and about 25%.
- 49) The method of any one of claims 46-49, wherein a G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%.
- 50) The method of any one of claims 46-49, wherein the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and 74%.
- 51) The method of claim 46, wherein preparing the culture medium comprises measuring Mn concentration in one or more batches of raw material components used to prepare the culture medium and selecting those one or more batches of raw material components that in combination contain about 8.5 ppb or less Mn.
- 52) The method of claim 51, wherein the culture medium is prepared to comprise between about 4.0 ppb and about 8.5 ppb Mn.
- 53) The method of claim 52, wherein the culture medium is prepared to comprise between about 4.0 ppb and about 6.5 ppb Mn.
- 54) The method of claim 53, wherein the culture medium is prepared to comprise between about 5.0 ppb and about 6.5 ppb Mn.
- 55) The method of any one of claims 46-54, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 27%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 65% and about 74%, and the culture medium is prepared to comprise between about 4.0 ppb and about 8.5 ppb Mn.
- 56) The method of any one of claims 46-54, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 15% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is prepared to comprise between about 4.0 ppb and about 6.5 ppb Mn.
- 57) The method of any one of claims 46-54, wherein the G1F oligosaccharide content of the anti-CD38 antibody is between about 21% and about 25%, the G0F oligosaccharide content of the anti-CD38 antibody is between about 68% and about 74%, and the culture medium is prepared to comprise between about 5.0 ppb and about 6.5 ppb Mn.
- 58) The method of any one of claims 46-57, wherein the culture medium is a basal medium or a feed medium.
- 59) The method of any one of claims 46-58, wherein culturing comprises a fed batch culture or a perfusion culture.
- 60) The method of any one of claims 46-59, wherein the host cell is an eukaryotic cell.

- 61) The method of claim 60, wherein the eukaryotic cell is a CHO cell, a PER.C6 cell, a NS0 cell, a Sp2/0 cell or a BHK cell. —
- 62) The method of claim 61, wherein the CHO cell is a CHO K1 cell, a CHO DG44 cell, a CHO S cell or a CHO DXB11 cell.
- 63) The method of claim 62, wherein the CHO cell is deficient in glutamine synthetase (GS).
- 64) The method of any one of claims 46-63, wherein the method is conducted under GMP-compliant conditions.
- 65) The method of any one of claims 46-64, wherein the anti-CD38 antibody comprises the VH of SEQ ID NO: 7 and the VL of SEQ ID NO: 9.
- 66) The method of any one of claims 46-65, wherein the anti-CD38 antibody comprises an IgG1 isotype.
- 67) The method of any one of claims 46-66, wherein the anti-CD38 antibody comprises the HC of SEQ ID NO: 9 and the LC of SEQ ID NO: 10.
- 68) The method of any one of claims 46-67, wherein the anti-CD38 antibody is a biosimilar.
- 69) The method of any one of claims 46-68, wherein formulating the drug product comprises formulating the anti-CD38 antibody at from about 20 mg/mL to about 180 mg/mL with recombinant human hyaluronidase (rHuPH20) in an amount of from about 30,000 U to about 45,000 U, histidine at a concentration of from about 5 mM to about 15 mM, sorbitol at a concentration of from about 100 mM to about 300 mM, PS-20 at a concentration of from about 0.01% w/v to about 0.04 % w/v and methionine at a concentration of from about 1 mg/mL to about 2 mg/mL, at a pH of about 5.6.
- 70) The method of any one of claims 46-69, wherein formulating the drug product comprises formulating the anti-CD38 antibody at about 120 mg/mL in about 2,000 U/ml recombinant human hyaluronidase (rHuPH20), about 5 mM to about 15 mM histidine, about 100 mM to about 300 mM sorbitol, about 0.01% w/v to about 0.04 % w/v PS-20 and about 1 mg/mL to about 2 mg/mL methionine, at a pH of about 5.6.
- 71) The method of any one of claims 46-68, wherein formulating the drug product comprises formulating the anti-CD38 antibody at 20 mg/ml in about 25 mM acetic acid, about 60 mM sodium chloride, about 140 mM mannitol and about 0.04% w/v polysorbate-20 (PS-20); at pH about 5.5.