

CLAIMS:

1. An isolated multispecific antibody comprising:

a first binding moiety that has binding specificity for CD3, comprising:

a first heavy chain polypeptide subunit comprising a heavy chain variable domain comprising a CDR1 sequence comprising a sequence of SEQ ID NO: 36, a CDR2 sequence comprising a sequence of SEQ ID NO: 37, and a CDR3 sequence comprising a sequence of SEQ ID NO: 38; and

a first light chain polypeptide subunit comprising a light chain variable domain comprising a CDR1 sequence comprising a sequence of SEQ ID NO: 39, a CDR2 sequence comprising a sequence of SEQ ID NO: 40, and a CDR3 sequence comprising a sequence of SEQ ID NO: 41;

a second binding moiety that has binding specificity for CD19, comprising:

a ~~second~~^{first} heavy chain polypeptide subunit comprising a heavy chain variable domain; wherein the first heavy chain polypeptide subunit further comprises a mutated human IgG4 constant region comprising mutations S228P, F234A, L235A, and T366W; and the

a second heavy chain polypeptide subunit comprising esing a mutated human IgG4 constant region comprising mutations S228P, F234A, L235A, T366S, L368A, and Y407V.

2. The isolated multispecific antibody of claim 1, wherein the mutated human IgG4 constant region of the ~~first heavy chain polypeptide subunit or the mutated human IgG4 constant region of~~ second heavy chain polypeptide subunit lacks a CH1 domain.

3. The isolated multispecific antibody of claim 1 or 2, wherein the mutated human IgG4 constant region of the first heavy chain polypeptide subunit comprises a sequence of SEQ ID NO: 73 or 55, and the mutated human IgG4 constant region of the second heavy chain polypeptide subunit comprises a sequence of SEQ ID NO: 72 or 54.

~~4. The isolated multispecific antibody of any one of claims 1-3, further comprising a first binding moiety that has binding specificity for CD3, comprising:~~

~~a heavy chain variable domain comprising a CDR1 sequence comprising a sequence of SEQ ID NO: 36, a CDR2 sequence comprising a sequence of SEQ ID NO: 37, and a CDR3 sequence comprising a sequence of SEQ ID NO: 38; and~~

~~a light chain variable domain comprising a CDR1 sequence comprising a sequence of SEQ ID NO: 39, a CDR2 sequence comprising a sequence of SEQ ID NO: 40, and a CDR3 sequence comprising a sequence of SEQ ID NO: 41.~~

~~5. The isolated multispecific antibody of claim 4, wherein:~~

~~the CDR1, CDR2 and CDR3 sequences in the heavy chain variable domain of the first binding moiety are present in a human VH framework; and~~

~~the CDR1, CDR2 and CDR3 sequences in the light chain variable domain of the first binding moiety are present in a human Vkappa framework.~~

~~64.~~ The isolated multispecific antibody of any one of claims 1-3~~claim 5~~, wherein:

the heavy chain variable domain of the first binding moiety comprises a sequence having at least 95% identity to SEQ ID NO: 42; and

the light chain variable domain of the first binding moiety comprises a sequence having at least 95% identity to SEQ ID NO: 43.

~~57.~~ The isolated multispecific antibody of claim 46, wherein:

the heavy chain variable domain of the first binding moiety comprises the sequence of SEQ ID NO: 42; and

the light chain variable domain of the first binding moiety comprises the sequence of SEQ ID NO: 43.

~~8. The isolated multispecific antibody of any one of claims 1-7, further comprising a second binding moiety having binding specificity to a protein other than CD3.~~

~~69.~~ The isolated multispecific antibody of any one of claims 1-5~~claim 8~~, wherein the second binding moiety comprises a single heavy chain variable region, in a monovalent or bivalent configuration.

~~10. The isolated multispecific antibody of claim 9, wherein the first binding moiety comprises a light chain polypeptide subunit and a heavy chain polypeptide subunit, and wherein the second binding moiety comprises a heavy chain polypeptide subunit.~~

~~744.~~ The isolated multispecific antibody of claim ~~640~~, wherein the first light chain polypeptide subunit of the first binding moiety comprises a light chain constant domain.

~~12. The isolated multispecific antibody of any one of claims 8-11, wherein the protein other than CD3 is a tumor-associated antigen (TAA) or a tumor-specific antigen (TSA).~~

~~13. The isolated multispecific antibody of claim 12, wherein the TAA is B-cell maturation antigen (BCMA).~~

~~14. The isolated multispecific antibody of claim 12, wherein the TAA is CD19.~~

~~15. The isolated multispecific antibody of claim 12, wherein the TAA is prostate specific membrane antigen (PSMA).~~

~~846.~~ A pharmaceutical composition comprising a multispecific antibody according to ~~any one of claims 1-15~~claim 1.

~~947.~~ A polynucleotide encoding a multispecific antibody according to any one of claims 1-~~745~~.

~~1048.~~ A vector comprising a polynucleotide according to claim ~~947~~.

~~119.~~ A cell comprising a vector according to claim ~~180~~.

~~1220.~~ A method of producing a multispecific antibody according to any one of claims 1-~~745~~, comprising growing a cell according to claim ~~119~~ under conditions permissive for expression of the multispecific antibody, and isolating the multispecific antibody from the cell.

~~21. — A method of treatment, comprising administering to an individual in need an effective dose of the multispecific antibody of any one of claims 1-15, or the pharmaceutical composition of claim 16.~~

~~22. — Use of a multispecific antibody of any one of claims 1-15 in the preparation of a medicament for the treatment of a disease or disorder in an individual in need.~~

1323. The multispecific antibody of any one of claims 1-715, or the pharmaceutical composition of claim 816, for use in therapy in an individual in need.

~~24. — A method for treating a disease or condition characterized by expression of BCMA, comprising administering to an individual in need an effective dose of the multispecific antibody according to claim 13, or a pharmaceutical composition comprising the multispecific antibody of claim 13.~~

~~25. — The method of claim 24, wherein the disease is an autoimmune disease.~~

~~26. — The method of claim 24, wherein the disease is a cancer.~~

~~27. — The method of claim 26, wherein the cancer is a myeloma.~~

~~28. — The method of claim 27, wherein the myeloma is multiple myeloma.~~

~~29. — A method for treating a disease or condition characterized by expression of PSMA, comprising administering to an individual in need an effective dose of the multispecific antibody according to claim 15, or a pharmaceutical composition comprising the multispecific antibody of claim 15.~~

~~30. — The method of claim 29, wherein the disease is a cancer.~~

~~31. The method of claim 29, wherein the cancer is prostate cancer.~~

~~1432. Use of the multispecific antibody according to any one of claims 1-7, or the pharmaceutical composition comprising the multispecific antibody of claim 8 in the manufacture of a medicament for~~A method for treating a disease or condition or disorder characterized by expression of CD19, wherein the multispecific antibody or the pharmaceutical composition comprising the multispecific antibody is to be administered to an individual in need in an effective dose~~comprising administering to an individual in need an effective dose of the multispecific antibody according to claim 14, or a pharmaceutical composition comprising the multispecific antibody of claim 14.~~

~~1533.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is diffuse large B-cell lymphoma (DLBCL).

~~1634.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is acute lymphoblastic leukemia (ALL).

~~1735.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is non-Hodgkin's lymphoma (NHL).

~~1836.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is systemic lupus erythematosus (SLE).

~~1937.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is rheumatoid arthritis (RA).

~~2038.~~ The ~~use~~method of claim ~~1432~~, wherein the disorder is multiple sclerosis (MS).

~~2139.~~ A kit for treating a disease or disorder in an individual in need, comprising a multispecific antibody of any one of claims 1-~~7~~~~15~~, or the pharmaceutical composition of claim ~~8~~~~16~~, and instructions for use.

~~2240.~~ The kit of claim ~~2139~~, further comprising at least one additional reagent.

2341. The kit of claim 2240, wherein the at least one additional reagent comprises a chemotherapeutic drug.

2442. A bispecific three-chain antibody like molecule that binds to human CD3 and human CD19, comprising:

(i) a first polypeptide subunit comprising the amino acid sequence of SEQ ID NO: 49;
(ii) a second polypeptide subunit comprising a sequence of SEQ ID NO: 56; wherein the first and second polypeptide subunits together form a first binding moiety that binds to human CD3; and

(iii) a third polypeptide subunit that binds to human CD19, comprising the amino acid sequence of SEQ ID NO: 67

~~a light chain variable domain (VL) comprising a sequence of SEQ ID NO: 43; and
a light chain constant domain (CL);
a second polypeptide subunit comprising:
a heavy chain variable domain (VH) comprising a sequence of SEQ ID NO: 42; and
a heavy chain constant domain (CH) comprising a sequence of SEQ ID NO: 72 or 73;
wherein the light chain variable domain and the heavy chain variable domain together form
a first binding moiety that binds to CD3; and
—— a third polypeptide subunit comprising:
a heavy chain only variable region, in a monovalent or bivalent configuration, that binds
to a protein other than CD3; and
a heavy chain constant domain (CH) comprising a sequence of SEQ ID NO: 54 or 55.~~

2543. The bispecific three-chain antibody like molecule of claim 2442, wherein the first polypeptide subunit comprises a light chain~~third polypeptide subunit comprises a heavy chain only variable region in a bivalent configuration that binds to BCMA.~~

26. The bispecific three-chain antibody like molecule of claim 24, wherein the second polypeptide subunit comprises a heavy chain.

27. The bispecific three-chain antibody like molecule of claim 24, wherein the third polypeptide subunit comprises a monovalent heavy chain.

28. The bispecific three-chain antibody like molecule of claim 27, wherein the monovalent heavy chain comprises a heavy chain-only variable region in a monovalent configuration.

29. The bispecific three-chain antibody like molecule of claim 27, wherein the monovalent heavy chain lacks a CH1 domain.

30. A bispecific three-chain antibody like molecule that binds to human CD3 and human CD19, comprising:

(i) a first polypeptide subunit comprising the amino acid sequence of SEQ ID NO: 49;

(ii) a second polypeptide subunit comprising the amino acid sequence of SEQ ID NO: 75, wherein the first and second polypeptide subunits together form a first binding moiety that

binds to human CD3; and

(iii) a third polypeptide subunit that binds to human CD19, comprising the amino acid sequence of SEQ ID NO: 78.

31. The bispecific three-chain antibody like molecule of claim 30, wherein the first polypeptide subunit comprises a light chain.

32. The bispecific three-chain antibody like molecule of claim 30, wherein the second polypeptide subunit comprises a heavy chain.

33. The bispecific three-chain antibody like molecule of claim 30, wherein the third polypeptide subunit comprises a monovalent heavy chain.

34. The bispecific three-chain antibody like molecule of claim 33, wherein the monovalent heavy chain comprises a heavy chain-only variable region in a monovalent configuration.

35. The bispecific three-chain antibody like molecule of claim 33, wherein the monovalent heavy chain lacks a CH1 domain.

36. A human, monoclonal IgG4 bispecific antibody comprising:

(i) a first binding arm that binds to human CD3; and

(ii) a second binding arm that binds to human CD19,

wherein the first binding arm comprises a first heavy chain comprising the amino acid sequence of SEQ ID NO: 56 and a light chain comprising the amino acid sequence of SEQ ID NO: 49; and

wherein the second binding arm comprises a monovalent second heavy chain comprising the amino acid sequence of SEQ ID NO: 67.

37. The human, monoclonal IgG4 bispecific antibody of claim 36, wherein the second binding arm does not comprise a light chain.

38. The human, monoclonal IgG4 bispecific antibody of claim 36, wherein the monovalent second heavy chain comprises a heavy chain-only variable region in a monovalent configuration.

39. The human, monoclonal IgG4 bispecific antibody of claim 36, wherein the monovalent second heavy chain lacks a CH1 domain.

40. A human, monoclonal IgG4 bispecific antibody comprising:

(i) a first binding arm that binds to human CD3; and

(ii) a second binding arm that binds to human CD19,

wherein the first binding arm comprises a first heavy chain comprising the amino acid sequence of SEQ ID NO: 75 and a light chain comprising the amino acid sequence of SEQ ID NO: 49; and

wherein the second binding arm comprises a monovalent second heavy chain comprising The amino acid sequence of SEQ ID NO: 78.

41. The human, monoclonal IgG4 bispecific antibody of claim 40, wherein the second binding arm does not comprise a light chain.

42. The human, monoclonal IgG4 bispecific antibody of claim 40, wherein the monovalent second heavy chain comprises a heavy chain-only variable region in a monovalent configuration.

43. The human, monoclonal IgG4 bispecific antibody of claim 40, wherein the monovalent second heavy chain lacks a CH1 domain.

~~44. The bispecific three-chain antibody like molecule of claim 43, comprising:
a first polypeptide subunit comprising a sequence of SEQ ID NO: 49;
a second polypeptide subunit comprising a sequence of SEQ ID NO: 56; and
a third polypeptide subunit comprising a sequence of SEQ ID NO: 58.~~

~~45. A pharmaceutical composition comprising the bispecific three-chain antibody like molecule of any one of claims 42-44.~~

~~46. A polynucleotide encoding a bispecific three-chain antibody like molecule according to any one of claims 42-44.~~

~~47. A vector comprising a polynucleotide according to claim 46.~~

~~48. A cell comprising a vector according to claim 47.~~

~~49. A method of producing a bispecific three-chain antibody like molecule according to any one of claims 42-44, comprising growing a cell according to claim 48 under conditions permissive for expression of the bispecific three-chain antibody like molecule, and isolating the bispecific three-chain antibody like molecule from the cell.~~

~~50. A method of treatment, comprising administering to an individual in need an effective dose of the bispecific three-chain antibody like molecule of any one of claims 42-44, or the pharmaceutical composition of claim 45.~~

~~51. — Use of a bispecific three-chain antibody like molecule of any one of claims 42-44 in the preparation of a medicament for the treatment of a disease or disorder in an individual in need.~~

~~52. — The bispecific three-chain antibody like molecule of any one of claims 42-44, or the pharmaceutical composition of claim 45, for use in therapy in an individual in need.~~

~~53. — A method for treating a disease or condition characterized by expression of BCMA, comprising administering to an individual in need an effective dose of the bispecific three-chain antibody like molecule according to any one of claims 42-44, or the pharmaceutical composition of claim 45.~~

~~54. — The method of claim 53, wherein the disease is an autoimmune disease.~~

~~55. — The method of claim 53, wherein the disease is a cancer.~~

~~56. — The method of claim 55, wherein the cancer is a myeloma.~~

~~57. — The method of claim 56, wherein the myeloma is multiple myeloma.~~

~~58. — A kit for treating a disease or disorder in an individual in need, comprising a bispecific three-chain antibody like molecule of any one of claims 42-44, or the pharmaceutical composition of claim 45, and instructions for use.~~

~~59. — The kit of claim 58, further comprising at least one additional reagent.~~

~~60. — The kit of claim 59, wherein the at least one additional reagent comprises a chemotherapeutic drug.~~