

WHAT IS CLAIMED:

1. An anti-CD19 single domain antibody (sdAb) comprising:

- (i) a CDR1 comprising the amino acid sequence of SEQ ID NO: 1; a CDR2 comprising the amino acid sequence of SEQ ID NO: 8; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 15;
- (ii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 22 or 108; a CDR2 comprising the amino acid sequence of SEQ ID NO: 29; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 36;
- (iii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 2; a CDR2 comprising the amino acid sequence of SEQ ID NO: 9; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 16;
- (iv) a CDR1 comprising the amino acid sequence of SEQ ID NO: 23 or 109; a CDR2 comprising the amino acid sequence of SEQ ID NO: 30; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 37;
- (v) a CDR1 comprising the amino acid sequence of SEQ ID NO: 3; a CDR2 comprising the amino acid sequence of SEQ ID NO: 10; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 17;
- (vi) a CDR1 comprising the amino acid sequence of SEQ ID NO: 24 or 110; a CDR2 comprising the amino acid sequence of SEQ ID NO: 31; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 38;
- (vii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 4; a CDR2 comprising the amino acid sequence of SEQ ID NO: 11; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 18;
- (viii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 25 or 111; a CDR2 comprising the amino acid sequence of SEQ ID NO: 32; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 39;
- (ix) a CDR1 comprising the amino acid sequence of SEQ ID NO: 5; a CDR2 comprising the amino acid sequence of SEQ ID NO: 12; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 19;
- (x) a CDR1 comprising the amino acid sequence of SEQ ID NO: 26 or 112; a CDR2 comprising the amino acid sequence of SEQ ID NO: 33; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 40;
- (xi) a CDR1 comprising the amino acid sequence of SEQ ID NO: 6; a CDR2 comprising the amino acid sequence of SEQ ID NO: 13; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 20;

(xii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 27 or 113; a CDR2 comprising the amino acid sequence of SEQ ID NO: 34; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 41;

(xiii) a CDR1 comprising the amino acid sequence of SEQ ID NO: 7; a CDR2 comprising the amino acid sequence of SEQ ID NO: 14; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 21;

(xiv) a CDR1 comprising the amino acid sequence of SEQ ID NO: 28 or 114; a CDR2 comprising the amino acid sequence of SEQ ID NO: 35; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 42; or

(xv) a CDR1 comprising the amino acid sequence of SEQ ID NO: 1; a CDR2 comprising the amino acid sequence of SEQ ID NO: 8; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 50; or

(xvi) a CDR1 comprising the amino acid sequence of SEQ ID NO: 22 or 108; a CDR2 comprising the amino acid sequence of SEQ ID NO: 103; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 36.

2. An anti-CD19 single domain antibody (sdAb) comprising:

(i) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 43;

(ii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 44;

(iii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 45;

(iv) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 46;

(v) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 47;

(vi) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 48;

(vii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 49;

(viii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 51;

(ix) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 52;

(x) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1,

CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 53;

(xi) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 54;

(xii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 55;

(xiii) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 56; or

(xiv) a CDR1, a CDR2, and a CDR3 having the amino acid sequences of the CDR1, CDR2, and CDR3, respectively, as set forth in SEQ ID NO: 104.

3. The anti-CD19 sdAb of claim 2, wherein the CDR1, CDR2 or CDR3 are determined according to the Kabat numbering scheme, the IMGT numbering scheme, the AbM numbering scheme, the Chothia numbering scheme, the Contact numbering scheme, or a combination thereof.

4. The anti-CD19 sdAb of any one of claims 1 to 3, further comprising one or more FR regions as set forth in SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53, SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, and/or SEQ ID NO: 104.

5. The anti-CD19 sdAb of any one of claims 1 to 4, comprising the amino acid sequence of SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53, SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, or SEQ ID NO: 104.

6. The anti-CD19 sdAb of any one of claims 1 to 4, wherein anti-CD19 sdAb comprises or consists of an amino acid sequence having at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or more sequence identity with the sequence of SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53, SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, or SEQ ID NO: 104.

7. The anti-CD19 sdAb of claim 1 or claim 2, wherein anti-CD19 sdAb is a camelid sdAb.

8. The anti-CD19 sdAb of claim 1 or claim 2, wherein anti-CD19 sdAb is a humanized sdAb.

9. The anti-CD19 sdAb of any one of claims 1 to 8, wherein the anti-CD19 sdAb is genetically fused or chemically conjugated to an agent.

10. A chimeric antigen receptor (CAR), comprising:

(a) an extracellular antigen binding domain comprising the anti-CD19 sdAb of any

one of claims 1 to 9;

- (b) a transmembrane domain; and
- (c) an intracellular signaling domain.

11. The CAR of claim 10, wherein the extracellular antigen binding domain further comprises one or more additional antigen binding domain(s).

12. The CAR of claim 11, wherein the extracellular antigen binding domain further comprises one additional antigen binding domain.

13. The CAR of claim 11, wherein the extracellular antigen binding domain further comprises two additional antigen binding domains.

14. The CAR of any one of claims 11 to 13, wherein the one or more additional antigen binding domain(s) bind to one or more antigen(s) selected from a group consisting of CD20, CD22, CD33, CD38, BCMA, CS1, ROR1, GPC3, CD123, IL-13R, CD138, c-Met, EGFRvIII, GD-2, NY-ESO-1, MAGE A3, and glycolipid F77.

15. The CAR of any one of claims 10 to 14, wherein the transmembrane domain is derived from a molecule selected from a group consisting of CD8 α , CD4, CD28, CD137, CD80, CD86, CD152, and PD1.

16. The CAR of claim 15, wherein the transmembrane domain is derived from CD8 α .

17. The CAR of any one of claims 10 to 16, wherein the intracellular signaling domain comprises a primary intracellular signaling domain of an immune effector cell.

18. The CAR of claim 17, wherein the primary intracellular signaling domain is derived from CD3 ζ .

19. The CAR of claim 17 or claim 18, wherein the intracellular signaling domain further comprises a co-stimulatory signaling domain.

20. The CAR of claim 19, wherein the co-stimulatory signaling domain is derived from a co-stimulatory molecule selected from the group consisting of CD27, CD28, CD137, OX40, CD30, CD40, CD3, LFA-1, CD2, CD7, LIGHT, NKG2C, B7-H3, ligands of CD83 and combinations thereof.

21. The CAR of claim 20, wherein the co-stimulatory signaling domain is derived from CD137.

22. The CAR of any one of claims 10 to 21, further comprising a hinge domain located between the C-terminus of the extracellular antigen binding domain and the N-terminus of the transmembrane domain.

23. The CAR of claim 22, wherein the hinge domain is derived from CD8 α .
24. The CAR of any one of claims 10 to 23, further comprising a signal peptide located at the N-terminus of the polypeptide.
25. The CAR of claim 24, wherein the signal peptide is derived from CD8 α .
26. A chimeric antigen receptor (CAR), comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 57, SEQ ID NO: 58, SEQ ID NO: 59, SEQ ID NO: 60, SEQ ID NO: 61, SEQ ID NO: 62, SEQ ID NO: 63, and SEQ ID NO: 105.
27. An isolated nucleic acid comprising a nucleic acid sequence encoding the anti-CD19 sdAb of any one of claims 1 to 9.
28. A vector comprising the isolated nucleic acid of claim 27.
29. An isolated nucleic acid comprising a nucleic acid sequence encoding the CAR of any one of claims 10 to 26.
30. A vector comprising the isolated nucleic acid of claim 29.
31. An engineered immune effector cell, comprising the CAR of any one of claims 10 to 26, the isolated nucleic acid of claim 29, or the vector of claim 30.
32. The engineered immune effector cell of claim 31, wherein the immune effector cell is a T cell or B cell.
33. A pharmaceutical composition, comprising the anti-CD19 sdAb of any one of claims 1 to 9, the engineered immune effector cell of claim 31 or claim 32, or the vector of claim 28 or claim 30, and a pharmaceutically acceptable excipient.
34. ~~A method of treating a disease or disorder in a subject, comprising administering to the subject an effective amount~~ Use of the anti-CD19 sdAb of any one of claims 1 to 9, the engineered immune effector cell of claim 31 or claim 32, or the pharmaceutical composition of claim 33 in the manufacture of a medicament for treating a disease or disorder in a subject.
35. The ~~use method~~ of claim 34, wherein the disease or disorder is a B cell associated disease or disorder and/or CD19 associated disease or disorder.
36. The ~~use method~~ of claim 35, wherein the disease or disorder is cancer.
37. The ~~use method~~ of claim 36, wherein the disease or disorder is a B cell malignancy.
38. The ~~use method~~ of claim 37, wherein the B cell malignancy is a B cell leukemia or B cell lymphoma.
39. The ~~use method~~ of claim 34, wherein the disease or disorder is selected from a group

consisting of marginal zone lymphoma (*e.g.*, splenic marginal zone lymphoma), diffuse large B cell lymphoma (DLBCL), mantle cell lymphoma (MCL), primary central nervous system (CNS) lymphoma, primary mediastinal B cell lymphoma (PMBL), small lymphocytic lymphoma (SLL), B cell prolymphocytic leukemia (B-PLL), follicular lymphoma (FL), burkitt lymphoma, primary intraocular lymphoma, chronic lymphocytic leukemia (CLL), acute lymphoblastic leukemia (ALL), hairy cell leukemia (HCL), precursor B lymphoblastic leukemia, non-hodgkin lymphoma (NHL), high-grade B-cell lymphoma (HGBL), and multiple myeloma (MM).

40. The ~~usemethod~~ of claim 34, wherein the disease or disorder is an autoimmune and/or inflammatory disease.

41. The ~~usemethod~~ of claim 40, wherein the autoimmune and/or inflammatory disease is associated with inappropriate or enhanced B cell numbers and/or activation.