improved in vivo therapeutic activity in clinical applications of chimeric receptor-modified T cells.

There is a need to identify methods for determining elements of chimeric receptor design that are important for therapeutic activity and cell populations to genetically modify and adoptively transfer that provide enhanced survival and efficacy in vivo.

Summary

In one example, the present disclosure refers to a nucleic acid encoding a chimeric receptor, the nucleic acid comprising: a) a polynucleotide encoding a ligand binding domain consisting of a single chain variable fragment (scFv) that binds to a CD19; b) a polynucleotide encoding a transmembrane domain; c) a polynucleotide encoding a polypeptide spacer located between the ligand binding domain and the transmembrane domain, wherein the polypeptide spacer is about 15 amino acids or less in length and comprises an amino acid sequence of X_1PPX_2P, wherein X_1 is a cysteine, glycine, or arginine and X_2 is a cysteine or a threonine (SEQ ID NO:1); and d) a polynucleotide encoding an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In one aspect, the present disclosure refers to a nucleic acid encoding a chimeric receptor, the chimeric receptor comprising an extracellular domain consisting of a single chain variable fragment (scFv) and an extracellular polypeptide spacer, a transmembrane domain and an intracellular signaling domain, wherein: (a) the scFv binds to a CD19 and comprises a variable light chain (VL) domain comprising a CDRL1, a CDRL2, and a CDRL3 of the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a CDRH1, a CDRH2, and a CDRH3 of the amino acid sequence encoded by SEQ ID NO:3; (b) the polypeptide spacer is located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and (c) the intracellular signaling domain comprises a CD3ζ signaling domain and a costimulatory domain.

In another example, the present disclosure refers to a nucleic acid encoding a chimeric receptor, the nucleic acid comprising: a) a polynucleotide

encoding a ligand binding domain that binds to a CD19, wherein the ligand binding domain comprises a single chain variable fragment (scFv) that comprises: a variable light chain (VL) domain comprising a VL domain present in an amino acid sequence encoded by SEQ ID NO:3 or a sequence having at least 90% identity thereto, and a variable heavy chain (VH) domain comprising a VH domain present in an amino acid sequence encoded by SEQ ID NO:3 or a sequence having at least 90% identity thereto; a VL domain and a VH domain comprising the VL domain and the VH domain amino acid sequences, respectively, present in the amino acid sequence encoded by SEQ ID NO:3; or an amino acid sequence encoded by SEQ ID NO:3; b) a polynucleotide encoding a transmembrane domain; c) a polynucleotide encoding a polypeptide spacer located between the ligand binding domain and the transmembrane domain, wherein the polypeptide spacer is about 15 amino acids or less in length and comprises an amino acid sequence of X_1PPX_2P, wherein X_1 is a

cysteine, glycine, or arginine and X_2 is a cysteine or a threonine (SEQ ID NO:1); and d) a polynucleotide encoding an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In another aspect, the present disclosure refers to a nucleic acid encoding a chimeric receptor, the nucleic acid comprising: (a) a polynucleotide encoding a single chain variable fragment (scFv) that binds to a CD19, wherein the scFv comprises: a variable light chain (VL) domain comprising a VL domain present in the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a VH domain present in the amino acid sequence encoded by SEQ ID NO:3; or the amino acid sequence encoded by SEQ ID NO:3; (b) a polynucleotide encoding a transmembrane domain; (c) a polynucleotide encoding a polypeptide spacer located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and (d) a polynucleotide encoding an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In a further aspect, the present disclosure refers to a chimeric receptor polypeptide encoded by the nucleic acid as described herein.

In yet another example, the present disclosure refers to a chimeric receptor polypeptide, the chimeric receptor comprising: a) a ligand binding domain consisting of a single chain variable fragment (scFv) that binds to a CD19; b) a transmembrane domain; c) a polypeptide spacer located between the ligand binding domain and the transmembrane domain, wherein the polypeptide spacer is about 15 amino acids or less in length and comprises an amino acid sequence of X_1PPX_2P, wherein X_1 is a cysteine, glycine, or arginine and X_2 is a cysteine or a threonine (SEQ ID NO:1); and d) an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In yet another aspect, the present disclosure refers to a chimeric receptor polypeptide, the chimeric receptor comprising an extracellular domain consisting of a single chain variable fragment (scFv) and an extracellular polypeptide spacer, a transmembrane domain and an intracellular signaling domain, wherein: (a) the scFv

binds to a CD19 and comprises a variable light chain (VL) domain comprising a CDRL1, a CDRL2, and a CDRL3 of the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a CDRH1, a CDRH2, and a CDRH3 of the amino acid sequence encoded by SEQ ID NO:3; (b) the polypeptide spacer is located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and (c) the intracellular signaling domain comprises a CD3ζ signaling domain and a costimulatory domain.

In yet another example, the present disclosure refers to a chimeric receptor polypeptide, the chimeric receptor comprising: a) a ligand binding domain that binds to a CD19, wherein the ligand binding domain comprises a single chain variable fragment (scFv) that comprises: a variable light chain (VL) domain comprising a VL domain present in an amino acid sequence encoded by SEQ ID NO:3 or a sequence having at least 90% identity thereto, and a variable heavy chain (VH) domain comprising a VH domain present in an amino acid sequence encoded by SEQ ID NO:3 or a sequence having at least 90% identity thereto; a VL domain and a VH domain comprising the VL domain and the VH domain amino acid sequences, respectively, present in the amino acid sequence encoded by SEQ ID NO:3; or an amino acid sequence encoded by SEQ ID NO:3; b) a transmembrane domain; c) a polypeptide spacer located between the ligand binding domain and the transmembrane domain, wherein the polypeptide spacer is about 15 amino acids or less in length and comprises an amino acid sequence of X_1PPX_2P, wherein X_1 is a cysteine, glycine, or arginine and X_2 is a cysteine or a threonine (SEQ ID NO:1); and d) an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In yet another aspect, the present disclosure refers to a chimeric receptor polypeptide, the chimeric receptor comprising: (a) a single chain variable fragment (scFv) that binds to a CD19 and comprises: a variable light chain (VL) domain comprising a VL domain present in the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a VH domain present in the amino acid sequence encoded by SEQ ID NO:3; or the amino acid sequence encoded by SEQ ID NO:3; (b) a transmembrane domain; (c) a polypeptide spacer located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and (d) an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

In a further example, the present disclosure refers to a chimeric receptor polypeptide encoded by a nucleic acid construct comprising the sequence set forth in SEQ ID NO:10 or a sequence having at least 90% identity thereto. In yet another aspect, the present disclosure refers to a chimeric receptor polypeptide encoded by a nucleic acid construct comprising the sequence set forth in SEQ ID NO:10.

In yet another aspect, the present disclosure refers to an expression vector comprising the nucleic acid as described herein or a nucleic acid encoding the chimeric receptor polypeptide as described herein.

In yet another aspect, the present disclosure refers to an expression vector encoding the amino acid sequenceMLLLVTSLLLCELPHPAFLLIPDIQMTQTTSSLSASLGDRVTISCRAS QDISKYLNWYQQKPDGTVKLLIYHTSRLHSGVPSRFSGSGSGTDYSLTISNLE QEDIATYFCQQGNTLPYTFGGGTKLEITGSTSGSGKPGSGEGSTKGEVKLQE SGPGLVAPSQSLSVTCTVSGVSLPDYGVSWIRQPPRKGLEWLGVIWGSETTY YNSALKSRLTIIKDNSKSQVFLKMNSLQTDDTAIYYCAKHYYYGGSYAMDY WGQGTSVTVSSESKYGPPCPPCPMFWVLVVVGGVLACYSLLVTVAFIIFWV KRGRKKLLYIFKQPFMRPVQTTQEEDGCSCRFPEEEEGGCELRVKFSRSADA PAYQ

QGQNQLYNELNLGRREEYDVLDKRRGRDPEMGGKPRRKNPQEGLYNELQ
KDKMAEAYSEIGMKGERRRGKGHDGLYQGLSTATKDTYDALHMQALPPR
LEGGGEGRGSLLTCGDVEENPGPRMLLLVTSLLLCELPHPAFLLIPRKVCNGI
GIGEFKDSLSINATNIKHFKNCTSISGDLHILPVAFRGDSFTHTPPLDPQELDIL
KTVKEITGFLLIQAWPENRTDLHAFENLEIIRGRTKQHGQFSLAVVSLNITSL
GLRSLKEISDGDVIISGNKNLCYANTINWKKLFGTSGQKTKIISNRGENSCKA
TGQVCHALCSPEGCWGPEPRDCVSCRNVSRGRECVDKCNLLEGEPREFVEN
SECIQCHPECLPQAMNITCTGRGPDNCIQCAHYIDGPHCVKTCPAGVMGEN
NTLVWKYADAGHVCHLCHPNCTYGCTGPGLEGCPTNGPKIPSIATGMVGA
LLLLLVVALGIGLFM.

In yet another aspect, the present disclosure refers to a host cell comprising the nucleic acid as described herein, the chimeric receptor polypeptide as described herein, or the expression vector as described herein.

In yet another aspect, the present disclosure refers to a composition comprising the host cell as described herein in a pharmaceutically acceptable excipient.

In yet another aspect, the present disclosure refers to an in vitro method for preparing a host cell, comprising:

CLAIMS

1. A nucleic acid encoding a chimeric receptor, the chimeric receptor comprising an extracellular domain consisting of a single chain variable fragment (scFv) and an extracellular polypeptide spacer, a transmembrane domain and an intracellular signaling domain, wherein:

(a) the scFv binds to a CD19 and comprises a variable light chain (VL) domain comprising a CDRL1, a CDRL2, and a CDRL3 of the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a CDRH1, a CDRH2, and a CDRH3 of the amino acid sequence encoded by SEQ ID NO:3;

(b) the polypeptide spacer is located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and

(c) the intracellular signaling domain comprises a CD3ζ signaling domain and a costimulatory domain.

2. A nucleic acid encoding a chimeric receptor, the nucleic acid comprising:

(a) a polynucleotide encoding a single chain variable fragment (scFv) that binds to a CD19, wherein the scFv comprises:

a variable light chain (VL) domain comprising a VL domain present in the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a VH domain present in the amino acid sequence encoded by SEQ ID NO:3;

or

the amino acid sequence encoded by SEQ ID NO:3;

(b) a polynucleotide encoding a transmembrane domain;

(c) a polynucleotide encoding a polypeptide spacer located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and

(d) a polynucleotide encoding an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

3. The nucleic acid of claim 1 or 2, wherein the scFv comprises a variable heavy chain (VH) domain and a variable light chain (VL) domain separated by a peptide linker.

4. The nucleic acid of claim 3, wherein the peptide linker comprises the amino acid sequence set forth in SEQ ID NO:36.

5. The nucleic acid of claim 3 or 4, wherein the scFv comprises a linker, and has a VL-linker-VH orientation.

6. The nucleic acid of any one of claims 1-5, wherein the polypeptide spacer is 12 amino acids in length.

7. The nucleic acid of any one of claims 1-6, wherein the polypeptide spacer consists of the amino acid sequence set forth in SEQ ID NO:21.

8. The nucleic acid of any one of claims 1 and 3-7, wherein the scFv comprises a variable light chain (VL) domain having a least 90% sequence identity to the VL domain present in the amino acid sequence encoded by SEQ ID NO:3; and a variable heavy chain (VH) domain having at least 90% identity to the VH domain present in the amino acid sequence encoded by SEQ ID NO:3.

9. The nucleic acid of any one of claims 1 and 3-8, wherein the scFv comprises a variable light chain (VL) domain comprising a CDRL1 sequence of RASQDISKYLN, a CDRL2 sequence of SRLHSGV, a CDRL3 sequence of GNTLPYTFG, and a variable heavy chain (VH) domain comprising a CDRH1 sequence of DYGVS, a CDRH2 sequence of VIWGSETTYYNSALKS, and a CDRH3 sequence of YAMDYWG.

10. The nucleic acid of any one of claims 1 and 3-9, wherein the scFv comprises a VL domain comprising a VL domain present in the amino acid sequence encoded by SEQ ID NO:3, and a VH domain comprising a VH domain present in the amino acid sequence encoded by SEQ ID NO:3.

11. The nucleic acid of any one of claims 1-10, wherein the transmembrane domain comprises a transmembrane domain of a CD8 or of a CD28.

12. The nucleic acid of any one of claims 1-11, wherein the transmembrane domain comprises a transmembrane domain of a CD28 comprising the amino acid sequence encoded by SEQ ID NO:5, or the amino acid sequence MFWVLVVVGGVLACYSLLVTVAFIIFWV.

13. The nucleic acid of any one of claims 1-12, wherein the costimulatory domain comprises:

the signaling domain of a 4-1BB; or

the signaling domain of a CD28.

14. The nucleic acid of any one of claims 1-13, wherein:

the transmembrane domain comprises a transmembrane domain of a CD28; and

the costimulatory domain comprises all or a portion of a 4-1BB signaling domain or all or a portion of a CD28 signaling domain.

15. The nucleic acid of claim 13 or 14, wherein the 4-1BB signaling domain comprises amino acids 214-255 of SEQ ID NO:15 or comprises the amino acid sequence encoded by SEQ ID NO:6.

16. The nucleic acid of claim 13 or 14, wherein the CD28 signaling domain comprises amino acids 180-220 of SEQ ID NO:14 or comprises a modified version thereof comprising an LL → GG substitution located at positions 186-187 of SEQ ID NO:14.

17. The nucleic acid of any one of claims 1-16, wherein the CD3ζ signaling domain comprises the amino acid sequence encoded by SEQ ID NO:7.

18. The nucleic acid of any one of claims 1-17, wherein:

the scFv has a VL-linker-VH orientation;

the polypeptide spacer consists of the sequence set forth in SEQ ID NO:21;

the transmembrane domain comprises the amino acid sequence encoded by the polynucleotide sequence set forth in SEQ ID NO:5; and

the intracellular signaling domain comprises the amino acid sequence encoded by the polynucleotide sequence set forth in SEQ ID NO:6 and the amino acid sequence encoded by SEQ ID NO:7.

19. The nucleic acid of any one of claims 1, 3-7, and 11-18, wherein:

the scFv comprises a VL domain having at least 90% sequence identity to the VL domain sequence present in the amino acid sequence encoded by SEQ ID NO:3, and a VH domain having at least 90% sequence identity to the VH domain sequence present in the amino acid sequence encoded by SEQ ID NO:3;

the polypeptide spacer consists of the amino acid sequence set forth in SEQ ID NO:21;

the transmembrane domain comprises the amino acid sequence encoded by SEQ ID NO:5; and

the intracellular signaling domain comprises the amino acid sequence encoded by SEQ ID NO:6 and comprises the amino acid sequence encoded by SEQ ID NO:7.

20. The nucleic acid of any one of claims 1, 3-7, and 11-18, wherein:

the scFv comprises a VL domain having at least 90% sequence identity to the VL domain sequence present in the amino acid sequence encoded by SEQ ID NO:3, and a VH domain having at least 90% sequence identity to the VH domain sequence present in the amino acid sequence encoded by SEQ ID NO:3;

the polypeptide spacer consists of the amino acid sequence encoded by SEQ ID NO:4;

the transmembrane domain comprises the amino acid sequence encoded by SEQ ID NO:5; and

the intracellular signaling domain comprises the amino acid sequence encoded by SEQ ID NO:6 and the amino acid sequence encoded by SEQ ID NO:7.

21. The nucleic acid of any one of claims 1-20, wherein the scFv is encoded by the sequence set forth in SEQ ID NO:3.

22. The nucleic acid of any one of claims 1-21, wherein the nucleic acid comprises:

(a) the sequence set forth in SEQ ID NO:3 encoding the scFv;

(b) the sequence set forth in SEQ ID NO:4 encoding the polypeptide spacer;

(c) the sequence set forth in SEQ ID NO:5 encoding the transmembrane domain;

(d) the sequence set forth in SEQ ID NO:6 encoding the costimulatory domain; and

(e) the sequence set forth in SEQ ID NO:7 encoding the CD3ζ signaling domain.

23. The nucleic acid of any one of claims 1-22, wherein the chimeric receptor is encoded by a nucleic acid construct comprising the sequence set forth in SEQ ID NO:10 or a sequence having at least 90% identity thereto.

24. A chimeric receptor polypeptide encoded by the nucleic acid of any one of claims 1-23.

25. A chimeric receptor polypeptide, the chimeric receptor comprising an extracellular domain consisting of a single chain variable fragment (scFv) and an extracellular polypeptide spacer, a transmembrane domain and an intracellular signaling domain, wherein:

(a) the scFv binds to a CD19 and comprises a variable light chain (VL) domain comprising a CDRL1, a CDRL2, and a CDRL3 of the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a CDRH1, a CDRH2, and a CDRH3 of the amino acid sequence encoded by SEQ ID NO:3;

(b) the polypeptide spacer is located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and

(c) the intracellular signaling domain comprises a CD3ζ signaling domain and a costimulatory domain.

26. A chimeric receptor polypeptide, the chimeric receptor comprising:

(a) a single chain variable fragment (scFv) that binds to a CD19 and comprises:

a variable light chain (VL) domain comprising a VL domain present in the amino acid sequence encoded by SEQ ID NO:3, and a variable heavy chain (VH) domain comprising a VH domain present in the amino acid sequence encoded by SEQ ID NO:3;

or

the amino acid sequence encoded by SEQ ID NO:3;

(b) a transmembrane domain;

(c) a polypeptide spacer located between the scFv and the transmembrane domain, wherein the polypeptide spacer is 15 amino acids or less in length and comprises the amino acid sequence set forth in SEQ ID NO:21; and

(d) an intracellular signaling domain that comprises a CD3ζ signaling domain and a costimulatory domain.

27. The chimeric receptor polypeptide of any one of claims 24-26, wherein the chimeric receptor polypeptide comprises the sequence DIQMTQTTSSLSASLGDRVTISCRASQDISKYLNWYQQKPDGTVKLLIYHTSRLHSGVPSRFSGSGSGTDYSLTISNLEQE DIATYFCQQGNTLPYTFGGGTKLEITGSTSGSGKPGSGEGSTKGEVKLQESGPGLVAPSQSLSVTCTVSGVSLPDYGVSW IRQPPRKGLEWLGVIWGSETTYYNSALKSRLTIIKDNSKSQVFLKMNSLQTDDTAIYYCAKHYYYGGSYAMDYWGQGTS VTVSSESKYGPPCPPCPMFWVLVVVGGVLACYSLLVTVAFIIFWVKRGRKKLLYIFKQPFMRPVQTTQEEDGCSCRFPEE EEGGCELRVKFSRSADAPAYQQGQNQLYNELNLGRREEYDVLDKRRGRDPEMGGKPRRKNPQEGLYNELQKDKMAE AYSEIGMKGERRRGKGHDGLYQGLSTATKDTYDALHMQALPPR.

28. A chimeric receptor polypeptide encoded by a nucleic acid construct comprising the sequence set forth in SEQ ID NO:10.

29. An expression vector comprising the nucleic acid of any one of claims 1-23 or a nucleic acid encoding the chimeric receptor polypeptide of any one of claims 24-28.

30. The expression vector of claim 29, further comprising a polynucleotide encoding a marker sequence, wherein the polynucleotide encoding the marker sequence is operably linked in frame with the nucleic acid encoding the chimeric receptor.

31. The expression vector of claim 30, wherein the nucleic acid encoding the chimeric receptor and the polynucleotide encoding the marker sequence are separated by a polynucleotide encoding a cleavable linker.

32. The expression vector of claim 30 or 31, wherein the marker sequence is a truncated EGFR sequence.

33. The expression vector of claim 32, wherein the truncated EGFR sequence is encoded by the polynucleotide set forth in SEQ ID NO:9.

34. The expression vector of any one of claims 31-33, wherein the cleavable linker comprises a T2A peptide.

35. The expression vector of claim 34, wherein the T2A peptide is encoded by the polynucleotide set forth in SEQ ID NO:8.

36. An expression vector encoding the amino acid sequence
MLLLVTSLLLCELPHPAFLLIPDIQMTQTTSSLSASLGDRVTISCRASQDISKYLNWYQQKPDGTVKLLIYHTSRLHSGVPS
RFSGSGSGTDYSLTISNLEQEDIATYFCQQGNTLPYTFGGGTKLEITGSTSGSGKPGSGEGSTKGEVKLQESGPGLVAPSQ
SLSVTCTVSGVSLPDYGVSWIRQPPRKGLEWLGVIWGSETTYYNSALKSRLTIIKDNSKSQVFLKMNSLQTDDTAIYYCA
KHYYYGGSYAMDYWGQGTSVTVSSESKYGPPCPPCPMFWVLVVVGGVLACYSLLVTVAFIIFWVKRGRKKLLYIFKQPF
MRPVQTTQEEDGCSCRFPEEEEGGCELRVKFSRSADAPAYQQGQNQLYNELNLGRREEYDVLDKRRGRDPEMGGKP
RRKNPQEGLYNELQKDKMAEAYSEIGMKGERRRGKGHDGLYQGLSTATKDTYDALHMQALPPRLEGGGEGRGSLLTC
GDVEENPGPRMLLLVTSLLLCELPHPAFLLIPRKVCNGIGIGEFKDSLSINATNIKHFKNCTSISGDLHILPVAFRGDSFTHT
PPLDPQELDILKTVKEITGFLLIQAWPENRTDLHAFENLEIIRGRTKQHGQFSLAVVSLNITSLGLRSLKEISDGDVIISGNK
NLCYANTINWKKLFGTSGQKTKIISNRGENSCKATGQVCHALCSPEGCWGPEPRDCVSCRNVSRGRECVDKCNLLEGE
PREFVENSECIQCHPECLPQAMNITCTGRGPDNCIQCAHYIDGPHCVKTCPAGVMGENNTLVWKYADAGHVCHLCH
PNCTYGCTGPGLEGCPTNGPKIPSIATGMVGALLLLLVVALGIGLFM.

37. A host cell comprising the nucleic acid of any one of claims 1-23, the chimeric receptor polypeptide of any one of claims 24-28, or the expression vector of any one of claims 29-36.

38. The host cell of claim 37, wherein the host cell is an immune cell.

39. The host cell of claim 37 or 38, wherein the host cell is a T cell.

40. The host cell of any one of claims 37-39, wherein the host cell is a CD8+ T cell or a CD4+ T cell.

41. The host cell of any one of claims 37-40, wherein the host cell is a CD8+ T cell selected from the group consisting of naïve CD8+ T cells, central memory CD8+ T cells, effector memory CD8+ T cells and bulk CD8+ T cells.

42. The host cell of any one of claims 37-40, wherein the host cell is a CD4+ T cell selected from the group consisting of naïve CD4+ T cells, central memory CD4+ T cells, effector memory CD4+ T cells, and bulk CD4+ T cells.

43. A composition comprising the host cell of any one of claims 37-42 in a pharmaceutically acceptable excipient.

44. The composition of claim 43, comprising a host cell of claims 41 and a host cell of claims 42.

45. The composition of claim 44, wherein the CD4+ T cells and CD8+ T cells are present at a 1:1 ratio in the composition.

46. An in vitro method for preparing a host cell, comprising:

introducing the nucleic acid of any one of claims 1-23 or the expression vector of any one of claims 29-36 into a T cell; and

culturing the cells in the presence of anti-CD3 or anti CD28 or both, and at least one homeostatic cytokine.

47. The method of claim 46, wherein the T cell is a CD8+ T cell or a CD4+ T cell.

48. Use of a pharmaceutical composition, comprising the chimeric receptor polypeptide of any one of claims 24-28, the host cell of any one of claims 37-42, or the composition of any one of claims 43-45 in the manufacture of a medicament for the treatment of a tumor expressing CD19.

49. Use of a population of immune cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28 in the manufacture of a medicament for treatment of a tumor expressing CD19.

50. The use of claim 49, wherein the population of immune cells comprise CD8+ T cells.

51. The use of claim 50, wherein the population of immune cells further comprises CD4+ T cells.

52. Use of a population of CD8+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28, in the manufacture of a medicament for treatment of a tumor expressing CD19.

53. The use of claim 52, wherein the population of CD8+ T cells is for use in combination with a population of CD4+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28.

54. Use of a population of CD4+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28, in the manufacture of a medicament for treatment of a tumor expressing CD19.

55. The use of claim 54, wherein the population of CD4+ T cells is for use in combination with a population of CD8+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28.

56. Use of a combination comprising a population of CD8+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28 and a population of CD4+ T cells comprising the nucleic acid of any one of claims 1-23 or the chimeric receptor polypeptide of any one of claims 24-28, in the manufacture of a medicament for treatment of a tumor expressing CD19.

57. The use of any one of claims 51, 53, 55, and 56, wherein the population of CD8+ T cells and the population of CD4+ T cells is for use at a ratio of 1:1 CD8+ T cells to CD4+ T cells.

58. The use of any one of claims 51, 53, and 55-57, wherein the population of CD8+ T cells and the population of CD4+ T cells express the same chimeric receptor.

59. The use of any one of claims 48-58, wherein the tumor is a solid tumor or hematologic malignancy.

60. The use of any one of claims 48-59, wherein the tumor is a lymphoma or a leukemia;
a B cell malignancy; or
a leukemia that is a chronic lymphocytic leukemia (CLL).

61. The use of any one of claims 48-60, wherein the tumor is a B cell malignancy.