CLAIMS

What is claimed is:

1. A polypeptide comprising (i) a transmembrane domain from CTLA4 or PD-1, (ii) an intracellular domain of an endogenous protein expressed on the surface of T cells that triggers the activation and/or proliferation of said T cells, and (iii) an extracellular domain that binds to an antigen, wherein

if the transmembrane domain is from CTLA4, the intracellular domain and extracellular domain of said polypeptide are not from CTLA4; and

if the transmembrane domain is from PD-1, the intracellular domain and extracellular domain of said polypeptide are not from PD-1.

2. The polypeptide of claim 1, wherein said polypeptide is a chimeric antigen receptor (CAR).

3. The polypeptide of claim 1 or claim 2, wherein a T cell expressing said polypeptide is activated or stimulated to proliferate when said polypeptide binds to said antigen.

4. The polypeptide of any of claims 1-3, wherein said polypeptide, when expressed on the surface of a T cell, directs the T cell to kill a cell expressing said antigen.

5. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is the polypeptide sequence encoded by exon 3 of a human CTLA4 gene.

6. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 1.

7. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 2.

8. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 3.

9. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 4.

10. The polypeptide of any of claims 1-4, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 5.

11. The polypeptide of any of claims 1-4, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO: 6.

12. The polypeptide of any of claims 1-4, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO: 7.

13. The polypeptide of any of claims 1-4, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO: 8.

14. The polypeptide of any of claims 1-13, wherein said extracellular domain comprises a receptor, or a portion of a receptor, that binds to said antigen.

15. The polypeptide of any of claims 1-13, wherein said extracellular domain is a receptor, or a portion of a receptor, that binds to said antigen.

16. The polypeptide of any of claims 1-13, wherein said extracellular domain comprises an antibody or an antigen-binding portion thereof.

17. The polypeptide of any of claims 1-13, wherein said extracellular domain is an antibody or an antigen-binding portion thereof.

18. The polypeptide of any of claims 1-13, wherein said extracellular domain comprises a single-chain Fv domain.

19. The polypeptide of claim 18, wherein said single-chain Fv domain comprises a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen.

20. The polypeptide of any of claims 1-19, wherein said antigen is an antigen on a tumor cell.

21. The polypeptide of claim 20, wherein said tumor cell is a cell in a solid tumor.

22. The polypeptide of claim 20, wherein said tumor cell is a cell of a blood cancer.

23. The polypeptide of any of claims 1-22, wherein said antigen is a tumor-associated antigen or a tumor-specific antigen.

24. The polypeptide of claim 23, wherein said tumor-associated antigen or tumor-specific antigen is Her2, prostate stem cell antigen (PSCA), alpha-fetoprotein (AFP), carcinoembryonic antigen (CEA), cancer antigen-125 (CA-125), CA19-9, calretinin, MUC-1, epithelial membrane protein (EMA), epithelial tumor antigen (ETA), tyrosinase, melanoma-associated antigen (MAGE), CD34, CD45, CD99, CD117, chromogranin, cytokeratin, desmin, glial fibrillary acidic protein (GFAP), gross cystic disease fluid protein (GCDFP-15), HMB-45 antigen, protein melan-A (melanoma antigen recognized by T lymphocytes; MART-1), myo-D1, muscle-specific actin (MSA), neurofilament, neuron-specific enolase (NSE), placental alkaline phosphatase, synaptophysis, thyroglobulin, thyroid transcription factor-1, the dimeric form of the pyruvate kinase isoenzyme type M2 (tumor M2-PK), CD19, CD22, CD27, CD30, CD70, GD2

hinge polypeptide sequence; (iii) a PD-1 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

41. A T lymphocyte comprising a membrane-bound polypeptide, wherein said polypeptide comprises (i) a transmembrane domain from CTLA4 or PD-1 (Programmed Cell Death 1), (ii) an intracellular domain of an endogenous protein expressed on the surface of lymphocytes and that triggers the activation and/or proliferation of said lymphocytes, and (iii) an extracellular domain that binds to an antigen, wherein

if the transmembrane domain is from CTLA4, the intracellular domain and extracellular domain of said polypeptide are not from CTLA4; and

if the transmembrane domain is from PD-1, the intracellular domain and extracellular domain of said polypeptide are not from PD-1.

42. The T lymphocyte of claim 41, wherein said polypeptide is a chimeric antigen receptor (CAR).

43. The T lymphocyte of claim 41 or claim 42, wherein a T lymphocyte expressing said polypeptide is activated or stimulated to proliferate when said polypeptide binds to said antigen.

44. The T lymphocyte of any of claims 41-43, wherein said polypeptide, when expressed on the surface of the T lymphocyte, directs the T lymphocyte to kill a cell expressing said antigen.

45. The T lymphocyte of any of claims 41-44, wherein the CTLA4 transmembrane domain is the polypeptide sequence encoded by exon 3 of a human *CTLA4* gene.

46. The T lymphocyte of any of claims 41-44, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO: 1.

47. The T lymphocyte of any of claims 41-44, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO:2.

48. The T lymphocyte of any of claims 41-44, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO:3.

49. The T lymphocyte of any of claims 41-44, wherein the CTLA4 transmembrane domain is or comprises the amino acid sequence of SEQ ID NO:4.

50. The T lymphocyte of any of claims 41-44, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO:6.

51. The T lymphocyte of any of claims 41-44, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO:7.

52. The T lymphocyte of any of claims 41-44, wherein the PD-1 transmembrane domain is or comprises the polypeptide sequence of SEQ ID NO:8.

53. The T lymphocyte of any of claims 41-52, wherein said extracellular domain comprises a receptor, or a portion of a receptor, that binds to said antigen.

54. The T lymphocyte of any of claims 41-52, wherein said extracellular domain is a receptor, or a portion of a receptor, that binds to said antigen.

55. The T lymphocyte of any of claims 41-52, wherein said extracellular domain comprises an antibody or an antigen-binding portion thereof.

56. The T lymphocyte of any of claims 41-52, wherein said extracellular domain is an antibody or an antigen-binding portion thereof.

57. The T lymphocyte of any of claims 41-52, wherein said extracellular domain comprises a single-chain Fv domain.

58. The T lymphocyte of claim 57, wherein said single-chain Fv domain comprises a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen.

59. The T lymphocyte of any of claims 41-58, wherein said antigen is an antigen on a tumor cell.

60. The T lymphocyte of claim 59, wherein said tumor cell is a cell in a solid tumor.

61. The T lymphocyte of claim 59, wherein said tumor cell is a cell of a blood cancer.

62. The T lymphocyte of any of claims 41-61, wherein said antigen is a tumor-associated antigen or a tumor-specific antigen.

63. The T lymphocyte of claim 62, wherein said tumor-associated antigen or tumor-specific antigen is Her2, prostate stem cell antigen (PSCA), alpha-fetoprotein (AFP), carcinoembryonic antigen (CEA), cancer antigen-125 (CA-125), CA19-9, calretinin, MUC-1, epithelial membrane protein (EMA), epithelial tumor antigen (ETA), tyrosinase, melanoma-associated antigen (MAGE), CD34, CD45, CD99, CD117, chromogranin, cytokeratin, desmin, glial fibrillary acidic protein (GFAP), gross cystic disease fluid protein (GCDFP-15), HMB-45 antigen, protein melan-A (melanoma antigen recognized by T lymphocytes; MART-1), myo-D1, muscle-specific actin (MSA), neurofilament, neuron-specific enolase (NSE), placental alkaline phosphatase, synaptophysis, thyroglobulin, thyroid transcription factor-1, the dimeric form of the pyruvate kinase isoenzyme type M2 (tumor M2-PK), an abnormal ras protein, an abnormal p53 protein, CD19, CD22, CD27, CD30, CD70, GD2 (ganglioside G2), EGFRvIII (epidermal growth factor variant III), sperm protein 17 (Spl7), mesothelin, PAP (prostatic acid phosphatase), prostein, TARP (T cell receptor gamma alternate reading frame protein), Trp-p8, STEAP1 (six-transmembrane epithelial antigen of the prostate 1), an abnormal ras protein, an abnormal p53 protein,

integrin αvβ3 (CD61), galactin, K-Ras (V-Ki-ras2 Kirsten rat sarcoma viral oncogene), or Ral-B.

64. The T lymphocyte of any of claims 41-63, wherein said extracellular domain is joined to said transmembrane domain by a linker, spacer or hinge polypeptide sequence.

65. The T lymphocyte of claim 64, wherein said linker, spacer or hinge polypeptide sequence is from CD28.

66. The T lymphocyte of claim 64, wherein said linker, spacer or hinge polypeptide sequence is from CTLA4.

67. The T lymphocyte of claim 64, wherein said linker, spacer or hinge polypeptide sequence is from PD-1.

68. The T lymphocyte of any of claims 41-67, wherein said intracellular domain is an intracellular domain of a protein that is expressed on the surface of T cells and triggers activation and/or proliferation of said T cells.

69. The T lymphocyte of claim 68, wherein said intracellular domain is a CD3ζ intracellular signaling domain.

70. The T lymphocyte of claim 68, wherein said intracellular domain is from a lymphocyte receptor chain, a TCR/CD3 complex protein, an Fc receptor subunit or an IL-2 receptor subunit.

71. The T lymphocyte of any of claims 41-70, wherein said polypeptide additionally comprises one or more co-stimulatory domains.

72. The T lymphocyte of claim 71, wherein said one or more co-stimulatory domains comprises one or more of a co-stimulatory CD27 polypeptide sequence, a co-stimulatory CD28 polypeptide sequence, a co-stimulatory OX40 (CD 134) polypeptide sequence, a co-stimulatory 4-1BB (CD137) polypeptide sequence, or a co-stimulatory inducible T-cell costimulatory (ICOS) polypeptide sequence.

73. The T lymphocyte of any of claims 41-72, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) an antigen-binding domain; (ii) a CD28 or CTLA4 hinge polypeptide sequence; (iii) a CTLA4 or PD-1 transmembrane domain; (iv) a costimulatory domain; and (v) an intracellular signaling domain.

74. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a CD28 hinge polypeptide sequence; (iii) a CTLA4 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

75. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a CTLA4 hinge polypeptide sequence; (iii) a CTLA4 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

76. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a CD28 hinge polypeptide sequence; (iii) a PD-1 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

77. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a CTLA4 hinge polypeptide sequence; (iii) a PD-1 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

78. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a PD-1 hinge polypeptide sequence; (iii) a CTLA4 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.

79. The T lymphocyte of claim 73, wherein said polypeptide comprises, in order, from N-terminus to C-terminus: (i) a single-chain Fv domain comprising a V_L linked to V_H by a flexible linker, wherein said V_L and V_H are from an antibody that binds said antigen; (ii) a PD-1 hinge polypeptide sequence; (iii) a PD-1 transmembrane domain; (iv) a CD28 costimulatory domain; and (v) a CD3ζ intracellular signaling domain.