

TFP molecule are administered in combination with an agent that treats the disease associated with CD19 or BCMA.

[0039] In one aspect, an isolated nucleic acid molecule provided herein, an isolated polypeptide molecule provided herein, an isolated TFP provided herein, a complex provided herein, a vector provided herein, or a cell provided herein, is for use as a medicament.

[0040] In one aspect, provided herein is a method of treating a mammal having a disease associated with expression of CD19 or BCMA comprising administering to the mammal an effective amount of a TFP molecule provided herein, a cell provided herein, or a polypeptide molecule provided herein, wherein less cytokines are released in the mammal compared a mammal administered an effective amount of a T-cell expressing an anti-CD19 chimeric antigen receptor (CAR) or an anti-BCMA CAR.

[0040a] In one aspect, provided herein is a pharmaceutical composition comprising (I) a T cell from a human subject, wherein the T cell comprises a recombinant nucleic acid molecule encoding a T-cell receptor (TCR) fusion protein (TFP) comprising

(a) a TCR subunit comprising

(i) at least a portion of a TCR extracellular domain, and

(ii) a TCR intracellular domain comprising a stimulatory domain from an intracellular signaling domain of CD3 epsilon, CD3 gamma, CD3 delta, TCR alpha or TCR beta; and

(b) a human or humanized antibody domain comprising an antigen binding domain; wherein the TCR subunit and the antibody domain are operatively linked, and wherein the TFP incorporates into a TCR when expressed in a T-cell; and

(II) a pharmaceutically acceptable carrier.

[0040b] In one aspect, provided herein is a use of an effective amount of the pharmaceutical composition as disclosed herein in the manufacture of a medicament for treating a mammal having a disease.

INCORPORATION BY REFERENCE

[0041] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference.

BRIEF DESCRIPTION OF THE DRAWINGS

[0042] The novel features of the invention are set forth with particularity in the appended claims. A better understanding of the features and advantages of the present invention will be obtained by reference to the following detailed description that sets forth illustrative

embodiments, in which the principles of the invention are utilized, and the accompanying drawings of which:

[0043] FIG. 1 is a schematic illustration demonstrating the use of T-cell receptor fusion polypeptides (TFPs) of the invention. An exemplary TFP contains an anti-CD19 scFv and a full-length CD3 epsilon polypeptide fused via a $(G_4S)_3$ linker sequence. When produced by or introduced into a T-cell, the TFP associates with other polypeptides of the endogenous T-cell receptor (TCR) (shown to include two CD3 epsilon polypeptides, one CD3 gamma polypeptide, one CD3 delta polypeptide, two CD3 zeta polypeptides, one TCR alpha subunit and one TCR beta subunit, where the horizontal grey segment represents the plasma membrane) to form a reprogrammed TCR in which one or both of the endogenous CD3 epsilon polypeptides are substituted by the TFP.

[0044] FIG. 2A represents schematic illustrations demonstrating exemplary variations of reprogrammed T-cell receptor fusion polypeptides (TFPs) of the invention.

[0045] Figure 2A illustrates an exemplary reprogrammed TCR containing a TFP that contains an anti-CD19 scFv and a full-length TCR $V\alpha$ polypeptide fused via a $(G_4S)_3$ linker sequence.

CLAIMS

1. A pharmaceutical composition comprising
 - (I) a T cell from a human subject, wherein the T cell comprises a recombinant nucleic acid molecule encoding a T-cell receptor (TCR) fusion protein (TFP) comprising
 - (a) a TCR subunit comprising
 - (i) at least a portion of a TCR extracellular domain, and
 - (ii) a TCR intracellular domain comprising a stimulatory domain from an intracellular signaling domain of CD3 epsilon, CD3 gamma, CD3 delta, TCR alpha or TCR beta; and
 - (b) a human or humanized antibody domain comprising an antigen binding domain;wherein the TCR subunit and the antibody domain are operatively linked, and wherein the TFP incorporates into a TCR when expressed in a T-cell; and
 - (II) a pharmaceutically acceptable carrier.
2. The pharmaceutical composition of claim 1, wherein the antigen binding domain is an anti-CD19 binding domain or an anti-B-cell maturation antigen (BCMA) binding domain.
3. The pharmaceutical composition of claim 1 or 2, wherein the encoded antigen binding domain is connected to the TCR extracellular domain by a linker sequence, optionally, wherein the encoded linker sequence comprises (G₄S)_n, wherein n=1 to 4.
4. The pharmaceutical composition of any one of claims 1-3, wherein the TCR subunit comprises (i) a TCR extracellular domain, (ii) a TCR transmembrane domain, and (iii) a TCR intracellular domain, wherein at least two of (i), (ii), and (iii) are from the same TCR subunit, optionally, wherein (i), (ii), and (iii) are from the same TCR subunit.
5. The pharmaceutical composition of any one of claims 1-4, wherein the human or humanized antibody domain comprises a scFv or a single domain antibody, wherein the single domain antibody is optionally a V_H domain.
6. The pharmaceutical composition of any one of claims 1-5, wherein the encoded antigen binding domain comprises (i) a light chain (LC) CDR1, LC CDR2 and LC CDR3 of an anti-CD19 light chain binding domain amino acid sequence with 70-100% sequence identity to SEQ ID NO: 25, SEQ ID NO: 27 and SEQ ID NO: 29, respectively, and/or (ii) a heavy chain (HC) CDR1, HC CDR2 and HC CDR3 of an anti-CD19 heavy chain binding domain amino acid sequence with 70-100% sequence identity to SEQ ID NO: 31, SEQ ID NO: 33 and SEQ ID NO: 35, respectively.
7. The pharmaceutical composition of any one of claims 1-6, wherein the TFP includes an extracellular domain of a TCR subunit that comprises an extracellular domain or portion thereof of a protein selected from the group consisting of a TCR alpha chain, a TCR beta chain, a CD3 epsilon TCR subunit, a CD3 gamma TCR subunit, a CD3 delta TCR subunit, functional fragments thereof, and amino acid sequences thereof having at least one but not more than 20 modifications.

8. The pharmaceutical composition of any one of claims 1-7, wherein the TFP includes a transmembrane domain that comprises a transmembrane domain of a protein selected from the group consisting of a TCR alpha chain, a TCR beta chain, a CD3 epsilon TCR subunit, a CD3 gamma TCR subunit, a CD3 delta TCR subunit, functional fragments thereof, and amino acid sequences thereof having at least one but not more than 20 modifications.
9. The pharmaceutical composition of any one of claims 1-8, wherein the TFP includes a transmembrane domain that comprises a transmembrane domain of a protein selected from the group consisting of a TCR alpha chain, a TCR beta chain, a TCR zeta chain, a CD3 epsilon TCR subunit, a CD3 gamma TCR subunit, a CD3 delta TCR subunit, CD45, CD4, CD5, CD8, CD9, CD16, CD22, CD33, CD28, CD37, CD64, CD80, CD86, CD134, CD137, CD154, functional fragments thereof, and amino acid sequences thereof having at least one but not more than 20 modifications.
10. The pharmaceutical composition of any one of claims 1-9, wherein the recombinant nucleic acid molecule further comprises a sequence encoding a costimulatory domain, optionally, wherein the costimulatory domain is a functional signaling domain obtained from a protein selected from the group consisting of OX40, CD2, CD27, CD28, CD30, ICAM-1, LFA-1 (CD11a/CD18), ICOS (CD278), and 4-1BB (CD137), and amino acid sequences thereof having at least one but not more than 20 modifications thereto.
11. The pharmaceutical composition of any one of claims 1-10, wherein the TFP includes an immunoreceptor tyrosine-based activation motif (ITAM) of a TCR subunit that comprises an ITAM or portion thereof of a protein selected from the group consisting of CD3 zeta TCR subunit, CD3 epsilon TCR subunit, CD3 gamma TCR subunit, CD3 delta TCR subunit, TCR zeta chain, Fc epsilon receptor 1 chain, Fc epsilon receptor 2 chain, Fc gamma receptor 1 chain, Fc gamma receptor 2a chain, Fc gamma receptor 2b1 chain, Fc gamma receptor 2b2 chain, Fc gamma receptor 3a chain, Fc gamma receptor 3b chain, Fc beta receptor 1 chain, TYROBP (DAP12), CD5, CD16a, CD16b, CD22, CD23, CD32, CD64, CD79a, CD79b, CD89, CD278, CD66d, functional fragments thereof, and amino acid sequences thereof having at least one but not more than 20 modifications thereto.
12. The pharmaceutical composition of any one of claims 1-11, wherein the T cell is a primary T-cell.
13. The pharmaceutical composition of any one of claims 1-12, wherein the T cell is a CD8⁺ or CD4⁺ T cell.
14. The pharmaceutical composition of any one of claims 1-13, wherein the T cell further comprises a nucleic acid encoding at least a portion of an inhibitory molecule, wherein the at least a portion of an inhibitory molecule associates with a second polypeptide that comprises a positive signal from an intracellular signaling domain, optionally, wherein the inhibitory molecule comprise first polypeptide that comprises at least a portion of PD1 and a second polypeptide comprising a costimulatory domain and primary signaling domain.
15. The pharmaceutical composition of any one of claims 1-14, wherein the T cell forms a conjugate with a human cell expressing an antigen that specifically interacts with the antigen binding domain, production

- of IL-2 by the T cell is increased in the presence of a human cell expressing an antigen that specifically interacts with the antigen binding domain, or wherein the T cell exhibits increased cytotoxicity to a human cell expressing an antigen that specifically interacts with the antigen binding domain.
16. The pharmaceutical composition of any one of claims 1-15, wherein the TCR intracellular domain comprises a stimulatory domain from an intracellular signaling domain of CD3 epsilon, CD3 gamma or CD3 delta.
 17. Use of an effective amount of the pharmaceutical composition of any one of claims 1-16 in the manufacture of a medicament for treating a mammal having a disease.
 18. The use of claim 17, wherein the disease is selected from the group consisting of a proliferative disease, a cancer, a malignancy, myelodysplasia, a myelodysplastic syndrome, and a preleukemia.
 19. The use of claim 17, wherein the disease is a hematologic cancer selected from the group consisting of B-cell acute lymphoid leukemia (B-ALL), T-cell acute lymphoid leukemia (T-ALL), acute lymphoblastic leukemia (ALL); chronic myelogenous leukemia (CML), chronic lymphocytic leukemia (CLL), B cell prolymphocytic leukemia, blastic plasmacytoid dendritic cell neoplasm, Burkitt's lymphoma, diffuse large B cell lymphoma, follicular lymphoma, hairy cell leukemia, small cell-follicular lymphoma, large cell-follicular lymphoma, malignant lymphoproliferative conditions, MALT lymphoma, mantle cell lymphoma, Marginal zone lymphoma, multiple myeloma, myelodysplasia, myelodysplastic syndrome, non-Hodgkin's lymphoma, plasmablastic lymphoma, plasmacytoid dendritic cell neoplasm, Waldenstrom macroglobulinemia, preleukemia, a disease associated with CD19 or BCMA expression, and combinations thereof.
 20. The use of any one of claims 17-19, wherein the pharmaceutical composition is to be administered in combination with an agent that treats the disease associated with CD19 or BCMA.