CLAIMS

We claim:

1. A designed cytokine comprising alpha helices H1, H2, H3, and H4, wherein:

from an amino terminus to a carboxy terminus,

a first loop (L1) connects H1 and H4;

a second loop (L2) connects H4 and H2;

a third loop (L3) connects H2 and H3; and

wherein the designed cytokine binds to IL-2 receptor βγ (IL-2R βγ).

2. The designed cytokine of claim 1, wherein the designed cytokine comprises one or more of:

(a)
APTSSSTKKTQLQLEHLLLDLQMILNGINNMNADPELVEFLNRWITFCQSIISTGS
LEDLKHLQALEEELKPLEEVLNLAQSKNFHLNPRDLISNINVLVLELK;

(b) a sequence of SEQ ID NO: 1-350;

(c) a sequence having at least 70% identity to a sequence of (b); or

(d) a sequence of SEQ ID NO: 1-38 or 150-350.

3. The designed cytokine of claim 1 or 2, wherein the designed cytokine comprises the sequence of (i)
APTSSSTKKTQLQLEHLLLDLQMILNGINNMNADPELVEFLNRWITFCQSIISTGS
LEDLKHLQALEEELKPLEEVLNLAQSKNFHLRPRDLISNINVIVLELK,

(ii)
APTSSSTKKTQLQLEHLLLDLQMILNGINNMNADPELVEFLNRWITFCQSIISTLT
AGGSLSGDLKHLQNLSEELKPLEEVLNLAQSKNFHLRPRDLISNINVIVLELK, or

(iii)
APTSSSTKKTQLQLEHLLLDLQMILNGINNMNADPELVEFLNRWITFCQSIISTGS
VDPEELAKELQKLEEELKPLEEVLNLAQSKNFHLRPRDLISNINVIVLELK.

4. The designed cytokine of any one of claims 1-3, wherein the designed cytokine is operably linked to a targeting moiety; or wherein the designed cytokine comprises a targeting moiety.

5. The designed cytokine of any one of claims 1-3, wherein a fusion protein comprises the designed cytokine and a targeting moiety.

6. The designed cytokine of any one of claims 4-5, wherein the targeting moiety binds to a component of a tumor microenvironment (TME), or
wherein the targeting moiety binds to one or more of T-cell surface glycoprotein CD8 (also known as cluster of differentiation 8), Programmed cell death protein 1 (PD-1), Programmed death-ligand 1 (PD-L1; also known as cluster of differentiation 274 (CD274) or B7 homolog 1 (B7-H1) polypeptide), T-cell immunoreceptor with Ig and ITIM domains (TIGIT), Cytotoxic T-lymphocyte protein 4 (CTLA4), Lymphocyte activation gene 3 protein (LAG3), or T-cell immunoglobulin mucin receptor 3 (TIM3).

7. The designed cytokine of any one of claims 4-6, wherein the targeting moiety comprises an antibody, an antibody mimetic, or a functional fragment thereof or wherein the targeting moiety comprises one or more of a monoclonal antibody, an antigen-binding fraction (Fab), a single-chain variable fraction (scFv), a domain antibody, one or more of a heavy chain (VH) and a light chain (VL) domain of an immunoglobulin (Ig) polypeptide or gene encoding the same, a heavy-chain antibody (a VH or a VHH), a camelid or camelid-like structured antibody, and a nanobody.

8. The designed cytokine of any one of claims 1-7, wherein the designed cytokine is operably linked to a tether; or wherein the designed cytokine comprises a tether.

9. The designed cytokine of any one of claims 1-7, wherein a fusion protein comprises the designed cytokine and a tether.

10. The designed cytokine of claim 8 or 9, wherein the tether comprises one or more of a nucleic acid sequence, an amino acid sequence, and a small molecule, optionally

wherein the tether comprises a sequence isolated or derived from a transmembrane sequence, and optionally wherein the tether comprises the sequence PLFIPVAVMVTAFSGLAFIIWLARRLKKGKK.

11. The designed cytokine of any one of claims 1-10, wherein the designed cytokine is operably linked to a second cytokine or a second Designed Cytokine, optionally wherein the designed cytokine comprises a second cytokine or a second Designed Cytokine or wherein a fusion protein comprises the designed cytokine and a second cytokine or a second Designed Cytokine, optionally wherein the second Designed Cytokine comprises a sequence of one or more of SEQ ID NO: 1-38 or 150-350.

12. The designed cytokine of claim 11, wherein the second cytokine comprises a sequence isolated or derived from one or more of an IL-2 polypeptide, an IL-12 polypeptide, an IL-15 polypeptide, an IL-18 polypeptide, an IL-21 polypeptide, an IL-23 polypeptide, an interferon alpha polypeptide, an interferon beta polypeptide, an interferon gamma polypeptide, and an interferon omega polypeptide.

13. The designed cytokine of claim 12, wherein the designed cytokine comprises a first targeting moiety and the second cytokine comprises a second targeting moiety, optionally wherein the first targeting moiety and the second targeting moiety are identical, or wherein the first targeting moiety and the second targeting moiety are not identical.

14. The designed cytokine of claim 11, wherein the designed cytokine comprises a first targeting moiety and the second Designed Cytokine comprises a second targeting moiety, optionally wherein the first targeting moiety and the second targeting moiety are identical, or wherein the first targeting moiety and the second targeting moiety are not identical.

15. The designed cytokine of claim 12 or 13, wherein the designed cytokine comprises a first tether and the second cytokine or the second Designed Cytokine comprises a

second tether, optionally wherein the first tether and the second tether are identical, or wherein the first tether and the second tether are not identical.

16. A nucleic acid encoding the designed cytokine of any one of claims 1-15 or a fusion protein comprising the designed cytokine of any one of claims 1-15.

17. The nucleic acid of claim 16, further comprising a regulatory element capable of driving expression of the designed cytokine, and optionally wherein the regulatory element comprises a promoter, optionally wherein the promoter is inducible, optionally wherein the promoter comprises a minimal promoter, optionally wherein the minimal promoter comprises a sequence isolated or derived from one or more of minimal promoter-1 ("minP1"), YB-TATA and human beta globin, optionally wherein the minP1 comprises a sequence of AGAGGGTATATAAAAGCTCGACTTCCAG, optionally wherein the minimal promoter comprises the sequence of TAGAGGGTATATAATGGGGGCCACTAGTCTACTACCAGAAAGCTTGGTACC GAGCTCGGATCCAGCCACC or CTAGAGGGTATATAATGGGGGCCACTAGTCTACTACCAGAAAGCTTGGTAC CGAGCTCGGATCCAGCCACC, and optionally wherein the regulatory element (i) comprises a response element, and optionally wherein the response element comprises a repeated sequence, or (ii) comprises a noncoding or an untranslated sequence isolated or derived from one or more of NF AT, NFkB, REL, RELA, IRF2, GATA3 and ATF3, optionally wherein the regulatory element comprises a noncoding or an untranslated sequence isolated or derived from a GATA3 gene and, optionally, wherein the GATA3 sequence comprises GTTATCTCTCACGAGATCT, and optionally wherein the regulatory element comprises a noncoding or an untranslated sequence isolated or derived from RELA and, optionally, wherein the RELA sequence comprises GGGGATTTCCA.

18. A vector comprising a nucleic acid of any one of claims 16 or 17, optionally wherein the vector comprises an expression vector or wherein the vector comprises a delivery vector.

19. The vector of claim 18, wherein the vector further comprises a sequence encoding an exogenous receptor, optionally

wherein the exogenous receptor comprises an antigen binding moiety, a T Cell Receptor (TCR), or a chimeric antigen receptor (CAR); and optionally wherein the antigen is expressed on or secreted within one or more of a tumor cell, a cancer cell, a component of a TME, and a TME.

20. A cell comprising a designed cytokine of any one of claims 1-15, a nucleic acid or fusion protein of any one of claims 16 or 17, or a vector of any one of claims 18 or 19.

21. The cell of claim 20, wherein the cell is a mammalian cell, is a human cell, a primary cell, a cultured cell, an immortalized cell, an immune cell, a stem cell, or a precursor cell capable of producing the immune cell; optionally wherein the stem cell is a hematopoietic stem cell (HSC), an induced pluripotent stem cell (iPSC) or a dedifferentiated immune cell; optionally wherein the immune cell is a T lymphocyte (T cell), an alpha beta T cell, a gamma delta T cell, a B lymphocyte (B cell), a macrophage or a natural killer (NK) cell; and optionally wherein the cell is ex vivo or in vitro.

22. A composition comprising the designed cytokine of any one of claims 1-15, the nucleic acid or fusion protein of any one of claims 16 or 17, the vector of any one of claims 18 or 19, or the cell of any one of claims 20 or 21.

23. A pharmaceutical composition comprising one or more of (1) the designed cytokine of any one of claims 1-15, the nucleic acid or fusion protein of any one of claims 16 or 17, the vector of any one of claims 18 or 19, and the cell of any one of claims 20 or 21 and (2) a pharmaceutically acceptable carrier.

24. The designed cytokine of any one of claims 1-15, the nucleic acid or fusion protein of any one of claims 16 or 17, the vector of any one of claims 18 or 19, the cell of any one of claims 20 or 21 or the pharmaceutical composition of claim 23 for use in therapy.

25. Use of the designed cytokine of any one of claims 1-15, the nucleic acid or fusion protein of any one of claims 16 or 17, the vector of any one of claims 18 or 19, the cell of any one of claims 20 or 21 or the pharmaceutical composition of claim 23 in the manufacture of a medicament for treating a cancer or a subtype thereof, optionally wherein the cancer or the subtype thereof comprises a liquid cancer, a hematological cancer, or a solid cancer.