

CLAIMS

1. An antibody cytokine engrafted protein comprising:
 - (a) a heavy chain variable region (VH), comprising Complementarity Determining Regions (CDRs) HCDR1, HCDR2, HCDR3; and
 - (b) a light chain variable region (VL), comprising LCDR1, LCDR2, LCDR3; and
 - (c) a mutated Interleukin 2 (IL2) molecule engrafted into the HCDR1, wherein the HCDR1 engrafted with the mutated IL2 molecule comprises SEQ ID NO: 7, SEQ ID NO: 10, SEQ ID NO: 13, SEQ ID NO: 16, SEQ ID NO: 39, SEQ ID NO: 42, SEQ ID NO: 45, or SEQ ID NO: 48, and wherein the mutated IL2 molecule contains a mutation that reduces the affinity of the mutated IL2 molecule to a high affinity IL2 receptor.

2. An antibody cytokine engrafted protein comprising:
 - (a) a heavy chain variable region (VH), comprising Complementarity Determining Regions (CDRs) HCDR1, HCDR2, HCDR3; and
 - (b) a light chain variable region (VL), comprising LCDR1, LCDR2, LCDR3; and
 - (c) a mutated Interleukin 2 (IL2) molecule engrafted into the HCDR1, wherein the HCDR1 engrafted with the mutated IL2 molecule comprises SEQ ID NO: 7, SEQ ID NO: 10, SEQ ID NO: 13, SEQ ID NO: 16, SEQ ID NO: 39, SEQ ID NO: 42, SEQ ID NO: 45, or SEQ ID NO: 48,

wherein the antibody cytokine engrafted protein preferentially expands T effector cells over regulatory T cells.

3. An antibody cytokine engrafted protein comprising:
 - (a) a heavy chain variable region (VH), comprising Complementarity Determining Regions (CDRs) HCDR1, HCDR2, HCDR3; and
 - (b) a light chain variable region (VL), comprising LCDR1, LCDR2, LCDR3;
 - (c) a mutated Interleukin 2 (IL2) molecule engrafted into the HCDR1, wherein the HCDR1 engrafted with the mutated IL2 molecule comprises SEQ ID NO: 7, SEQ ID NO: 10, SEQ ID NO: 13, SEQ ID NO: 16, SEQ ID NO: 39, SEQ ID NO: 42, SEQ ID NO: 45, or SEQ ID NO: 48,

wherein the antibody cytokine engrafted protein stimulates one or more of CD8 T effector cell, NK cell proliferation, or combination thereof, greater than recombinant

IL2 or a modified aglycosylated IL2 lacking an N-terminal alanine and having a S125C mutation.

4. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the mutated IL2 molecule comprises SEQ ID NO: 4.
5. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the mutated IL2 molecule comprises SEQ ID NO: 6.
6. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the VH comprises SEQ ID NO: 19 or SEQ ID NO: 51.
7. The antibody cytokine engrafted protein of any one of claims 1 to 6, further comprising an IgG class antibody heavy chain.
8. The antibody cytokine engrafted protein of claim 7, wherein the IgG class heavy chain is selected from IgG1, IgG2, and IgG4.
9. The antibody cytokine engrafted protein of claim 7, wherein the IgG class heavy chain comprises SEQ ID NO: 21 or SEQ ID NO: 53.
10. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the IgG class light chain comprises SEQ ID NO:69 and the IgG class heavy chain comprises SEQ ID NO:53.
11. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the IgG class light chain comprises SEQ ID NO:37 and the IgG class heavy chain comprises SEQ ID NO:21.
12. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the IgG class light chain comprises SEQ ID NO:69 and the IgG class heavy chain comprises SEQ ID NO:21.

13. The antibody cytokine engrafted protein of any one of claims 1 to 3, wherein the IgG class light chain comprises SEQ ID NO:37 and the IgG class heavy chain comprises SEQ ID NO:53.
14. A pharmaceutical composition comprising the antibody cytokine engrafted protein of any one of claims 1 to 13 and a pharmaceutically acceptable carrier.
15. Use of the antibody cytokine engrafted protein of any one of claims 1 to 13 or the pharmaceutical composition of claim 14 in the manufacture of a medicament for the treatment of cancer.
16. The use of claim 15, wherein the cancer is selected from the group consisting of melanoma, lung cancer, colorectal cancer, prostate cancer, breast cancer, and lymphoma.
17. The use of claim 15 or 16 for combination therapy of cancer, wherein the antibody cytokine engrafted protein or pharmaceutical composition is to be administered in combination with a therapeutic agent.
18. The use of claim 17, wherein the therapeutic agent is a second antibody cytokine engrafted protein.
19. The use of claim 17, wherein the therapeutic agent is an immune checkpoint inhibitor.
20. The use of claim 19, wherein the immune checkpoint inhibitor is selected from the group consisting of: PD-1, PD-L1, PD-L2, TIM3, CTLA-4, LAG-3, CEACAM-1, CEACAM-5, VISTA, BTLA, TIGIT, LAIR1, CD160, 2B4 and TGFR.
21. The use of claim 17, wherein the therapeutic agent is a tyrosine kinase inhibitor.
22. A method of preferentially expanding T effector cells over regulatory T cells in a patient in need thereof, wherein a therapeutically effective amount of the antibody

cytokine engrafted protein of any one of claims 1 to 13 or the pharmaceutical composition of claim 14 is to be administered to the patient.

23. The method of claim 22, wherein the T effector cells are expanded and Treg cells are not expanded.

24. The method of any one of claims 22 to 23, wherein an immune checkpoint inhibitor is to be administered.

25. The method of claim 24, wherein the immune checkpoint inhibitor is selected from the group consisting of: PD-1, PD-L1, PD-L2, TIM3, CTLA-4, LAG-3, CEACAM-1, CEACAM-5, VISTA, BTLA, TIGIT, LAIR1, CD160, 2B4 and TGFR.

26. The antibody cytokine engrafted protein of any one of claims 1 to 13 or the pharmaceutical composition of claim 14 for use in the treatment of cancer.

27. An isolated nucleic acid encoding an antibody cytokine engrafted protein comprising: (i) a heavy chain of SEQ ID NO:22 and a light chain of SEQ ID NO:38; or (ii) a heavy chain of SEQ ID NO:54 and a light chain of SEQ ID NO:70.

28. A recombinant host cell suitable for the production of an antibody cytokine engrafted protein, comprising the nucleic acid of claim 27 encoding the heavy and light chain polypeptides of the antibody cytokine engrafted protein, and optionally, a secretion signal.

29. The recombinant host cell of claim 28, which is a mammalian cell line.

30. The recombinant host cell of claim 29, wherein the mammalian cell line is a CHO cell line, Cos cell line, HeLa cell line, myeloma cell line, transformed B-cell line, or a hybridoma.