

CLAIMS

1. An interleukin 2 (IL-2) mutein having:
 - (a) reduced binding affinity for interleukin 2 receptor γ (IL-2R γ) as compared to an IL-2 polypeptide encoded by SEQ ID NO: 2; and
 - (b) 15-95% $E_{\max}$ as compared to an IL-2 polypeptide encoded by SEQ ID NO: 2.
2. The IL-2 mutein of claim 1, wherein the mutein comprises:
 - (i) one or more amino acid substitutions that increase IL-2R β binding affinity compared to a polypeptide encoded by SEQ ID NO: 1, selected from L80F, R81D, L85V, I86V, and I92F, numbered in accordance with the amino acid sequence of SEQ ID NO: 1; and/or
 - (ii) one or more amino acid substitutions that reduce IL-2R γ receptor binding affinity and results in 15-95% of $E_{\max}$ compared to a polypeptide encoded by SEQ ID NO: 2, selected from (A) L18R and Q22E; and (B) the amino acid position 126, numbered in accordance with the amino acid sequence of SEQ ID NO: 2.
3. The IL-2 mutein of claim 2, wherein the amino acid substitution at position 126 of SEQ ID NO: 2 is selected from the group consisting of Q126A, Q126C, Q126D, Q126E, Q126G, Q126H, Q126I, Q126K, Q126M, Q126R, Q126S, or Q126T.
4. The IL-2 mutein of claim 3, wherein the mutein comprises the amino acid substitution Q126H, Q126K, or Q126M.
5. The IL-2 mutein of claim 3, wherein the mutein comprises an amino acid substitution Q126H.
6. An interleukin 2 (IL-2) mutein having (a) reduced binding affinity for interleukin 2 receptor γ (IL-2R γ); and (b) 15-95% $E_{\max}$ as compared to a polypeptide encoded by SEQ ID NO: 1.
7. The IL-2 mutein of claim 6, wherein the mutein comprises one or more amino acid substitutions that reduce IL-2R γ receptor binding affinity and results in 15-95% $E_{\max}$ as compared to a polypeptide encoded by SEQ ID NO: 1, selected from (A) L18R and Q22E; and (B) amino acid position 126, numbered in accordance with the amino acid sequence of SEQ ID NO: 1.

8. The IL-2 mutein of claim 7, wherein the mutein comprises an amino acid substitution at position 126 of SEQ ID NO: 1 selected from the group consisting of Q126A, Q126C, Q126D, Q126E, Q126G, Q126H, Q126I, Q126K, Q126M, Q126R, Q126S, or Q126T.
9. The IL-2 mutein of claim 8, wherein the mutein comprises an amino acid substitution Q126H, Q126K, or Q126M.
10. The IL-2 mutein of claim 9, wherein the mutein comprises an amino acid substitution Q126H.
11. The IL-2 mutein of any one of claims 1-10, wherein the mutein is structurally modified to increase half-life.
12. The IL-2 mutein of claim 11, wherein said modification comprises one or more modifications selected from the group consisting of fusion to a human Fc antibody fragment, fusion to albumin, and PEGylation.
13. The IL-2 mutein of any one of claims 1-12, wherein said mutein increases proliferation of regulatory T (T_{reg}) cells and/or induces minimal proliferation of potentially inflammatory T cells.
14. The IL-2 mutein of any one of claims 1-13, wherein said mutein causes expansion of T_{reg} cells and does not promote expansion of potentially inflammatory T cells and Natural Killer (NK) cell or granulocytes.
15. The IL-2 mutein of claim 14, wherein the mutein induces less proliferation of potentially inflammatory T cells compared to the polypeptide encoded by SEQ ID NO: 1 or SEQ ID NO: 2.
16. The IL-2 mutein of claim 15, wherein said potentially inflammatory T cells are $CD4^{+}IFN\gamma^{+}$ T cells or are $CD8^{+}IFN\gamma^{+}$ T cells.
17. The IL-2 mutein of any one of claims 1-16, wherein the mutein increases proliferation of T_{reg} cells by at least 3 fold and/or induces less $IFN\gamma$ secretion compared to the polypeptide encoded by SEQ ID NO: 1 or SEQ ID NO: 2.
18. The IL-2 mutein of any one of claims 1-17, wherein said mutein does not induce $IFN\gamma$ secretion from $CD8^{+}$ T cells and/or other inflammatory immune cell subsets.

19. The IL-2 mutein of any one of claims 1-18, wherein the mutein has 70-95% $E_{\max}$ as compared to the polypeptide encoded by SEQ ID NO: 1.

20. A nucleic acid encoding the IL-2 mutein of any one of claims 1-19.

21. A vector comprising the nucleic acid of claim 20.

22. A host cell comprising the nucleic acid of claim 20 or the vector of claim 21.

23. A sterile pharmaceutical composition comprising the IL-2 mutein of any one of claims 1-19 and a pharmaceutically acceptable excipient.

24. A syringe comprising (a) the IL-2 mutein of any one of claims 1-19; (b) the nucleic acid of claim 20; (c) the vector of claim 21; (d) the host cell of claim 22; and/or (e) the pharmaceutical composition of claim 23.

25. A catheter comprising (a) the IL-2 mutein of any one of claims 1-19; (b) the nucleic acid of claim 20; (c) the vector of claim 21; (d) the host cell of claim 22; and/or (e) the pharmaceutical composition of claim 23.

26. A kit comprising:

(a) one or more of (i) the IL-2 mutein of any one of claims 1-19; (ii) the nucleic acid of claim 20; (iii) the vector of claim 21; (iv) the host cell of claim 22; (v) the sterile pharmaceutical composition of claim 23; (vi) the syringe of claim 24; and/or (vii) the catheter of claim 25; and

(b) written instructions for using the (i) the IL-2 mutein of any one of claims 1-19; (ii) the nucleic acid of claim 20; (iii) the vector of claim 21; (iv) the host cell of claim 22; (v) the sterile pharmaceutical composition of claim 23; (vi) the syringe of claim 24; and/or (vii) the catheter of claim 25.

27. Use of the (a) a therapeutically effective amount of the IL-2 mutein of any one of claims 1-19; (b) the nucleic acid of claim 20; (c) the vector of claim 21; (d) the host cell of claim 22; and/or (e) the pharmaceutical composition of claim 23 in the preparation of a medicament for the treatment of an autoimmune disease in an individual in need thereof.

28. The use of claim 27, wherein the autoimmune disease is selected from the group consisting of rheumatoid arthritis, insulin-dependent diabetes mellitus, hemolytic anemias, rheumatic fever, thyroiditis, Crohn's disease, myasthenia gravis, glomerulonephritis, autoimmune

hepatitis, multiple sclerosis, alopecia areata, psoriasis, vitiligo, dystrophic epidermolysis bullosa, systemic lupus erythematosus, and graft vs. host disease.

29. The use of claim 28, wherein the autoimmune disease is graft vs. host disease.

30. The use of any one of claims 27-29, further comprising the IL-2 mutein of any one of claims 1-19 or the pharmaceutical composition of claim 23 to be administered in combination with an antibody that targets the mutein to a specific cell type.

31. The use of claim 30, wherein the cell type is a regulatory T (T_{reg}) cell.

32. The use of claim 30 or claim 31, wherein the antibody is covalently or non-covalently linked to the IL-2 mutein.

33. A method for producing the IL-2 mutein of any one of claims 1-19 comprising culturing the host cell of claim 22 under suitable conditions for the production of the mutein.

34. The method of claim 33 further comprising isolating and/or purifying the produced IL-2 mutein.

35. The method of claim 33 or 34, further comprising structurally modifying the IL-2 mutein to increase half-life.

36. The method of claim 35, wherein said modification comprises one or more alterations selected from the group consisting of fusion to a human Fc antibody fragment, fusion to albumin, and PEGylation.

37. Use of the IL-2 mutein of any one of claims 1-19 in the preparation of a composition for preventing the proliferation of potentially inflammatory T cells and/or preventing secretion of IFN γ from CD8⁺ T cells or other inflammatory immune cell subsets, said use comprising contacting the composition with a cell expressing an interleukin 2 receptor γ (IL-2R γ).

38. The use of claim 37, wherein said potentially inflammatory T cells are CD4⁺CD44⁺IFN γ ⁺ T cells or are CD8⁺CD44⁺IFN γ ⁺ T cells.

39. The use of claim 37 or claim 38, wherein the contacting is performed *in vitro*, *in vivo*, or *ex vivo*.

40. Use of an interleukin 2 (IL-2) mutein having: (i) reduced binding affinity for interleukin 2 receptor γ (IL-2R γ) as compared to the polypeptide encoded by SEQ ID NO: 2; and (ii) 0-50%

$E_{\max}$ as compared to the polypeptide encoded by SEQ ID NO: 2, in the preparation of a composition for decreasing proliferation of regulatory T (T_{reg}) cells, said use comprising contacting the composition with a T_{reg} cell.

41. The use of claim 40, wherein the IL-2 mutein comprises:

(i) one or more amino acid substitutions that increase IL-2R β binding affinity compared to the polypeptide encoded by SEQ ID NO: 1, selected from L80F, R81D, L85V, I86V, and I92F, numbered in accordance with the amino acid sequence of SEQ ID NO: 1; and

(ii) one or more amino acid substitutions that reduce IL-2R γ receptor binding affinity and results in 0-50% of the $E_{\max}$ compared to the polypeptide encoded by SEQ ID NO: 2, selected from (A) L18R and Q22E; and (B) the amino acid position 126, numbered in accordance with the amino acid sequence of SEQ ID NO: 2.

42. Use of an IL-2 mutein having: (i) reduced binding affinity for IL-2R γ as compared to the polypeptide encoded by SEQ ID NO: 1; and (ii) 0-50% $E_{\max}$ as compared to the polypeptide encoded by SEQ ID NO: 1, in the preparation of a composition for decreasing proliferation of regulatory T (T_{reg}) cells, said use comprising contacting the composition with a T_{reg} cell.

43. The use of claim 42, wherein the mutein comprises one or more amino acid substitutions that reduce IL-2R γ receptor binding affinity and results in 0-50% $E_{\max}$ as compared to a polypeptide encoded by SEQ ID NO: 1, selected from (A) L18R and Q22E; and (B) amino acid position 126, numbered in accordance with the amino acid sequence of SEQ ID NO: 1.

44. The use of claim 41 or claim 43, wherein the amino acid substitution at position 126 of SEQ ID NO: 1 or SEQ ID NO: 2 is selected from the group consisting of Q126A, Q126C, Q126D, Q126E, Q126G, Q126H, Q126I, Q126K, Q126M, Q126R, Q126S, or Q126T.

45. The use of claim 44, wherein the mutein comprises the amino acid substitution Q126H, Q126K, or Q126M.

46. The use of claim 45, wherein the mutein comprises an amino acid substitution Q126H.

47. The use of any one of claims 39-46, further comprising the muteins to be administered with one or more antibodies that target the mutein to a T_{reg} cell.

48. The use of claim 47, wherein the antibody is covalently or non-covalently linked to the mutein.

49. The use of any one of claims 39-48, wherein the contacting is performed *in vitro*, *in vivo*, or *ex vivo*.