

CLAIMS

1. A T cell wherein a transgene is integrated at a first site within the genome of the T cell such that expression of the transgene is under control of an endogenous promoter of the T cell, wherein the transgene encodes a therapeutic protein or therapeutic nucleic acid.

2. A T cell wherein a first transgene is integrated at a first site within the genome of the T cell such that expression of the first transgene is under control of a first endogenous promoter of the T cell, and wherein a second transgene is integrated at a second site within the genome of the T cell, such that expression of the second transgene is under the control of a second endogenous promoter, wherein said first and second endogenous promoters are different promoters, and wherein the first transgene encodes a first therapeutic protein or first therapeutic nucleic acid, and the second transgene encodes a second therapeutic protein or second therapeutic nucleic acid, preferably wherein the first therapeutic protein or first therapeutic nucleic acid is different from said second therapeutic protein or second therapeutic nucleic acid, respectively.

3. The T cell of claim 1 or 2, wherein the T cell is an immunostimulatory T cell.

4. The T cell of claim 1 or 2, wherein the T cell is an immunoinhibitory T cell.

5. An isolated population of T cells, which comprises a plurality of the T cell of claim 1 or 2.

6. An isolated population of T cells, which comprises a plurality of the T cell of claim 3.

7. An isolated population of T cells, which comprises a plurality of the T cell of claim 4.

8. A pharmaceutical composition comprising a therapeutically effective amount of the T cell of claim 1 or 2; and a pharmaceutically acceptable carrier.

9. A pharmaceutical composition comprising a therapeutically effective amount of a population of T cells, which population comprises a plurality of the T cell of claim 1 or 2; and a pharmaceutically acceptable carrier.

10. A pharmaceutical composition comprising a therapeutically effective amount of the T cell of claim 3; and a pharmaceutically acceptable carrier.

11. A pharmaceutical composition comprising a therapeutically effective amount of a population of T cells, which population comprises a plurality of the T cell of claim 3; and a pharmaceutically acceptable carrier.

12. A pharmaceutical composition comprising a therapeutically effective amount of the T cell of claim 4; and a pharmaceutically acceptable carrier.

13. A pharmaceutical composition comprising a therapeutically effective amount of a population of T cells, which population comprises a plurality of the T cell of claim 4; and a pharmaceutically acceptable carrier.

14. Use of a therapeutically effective amount of the T cell of claim 1 or 2 in the manufacture of a medicament for treating a subject with T cell therapy in need thereof, wherein the medicament is to be administered to the subject.

15. Use of a therapeutically effective amount of the T cell population of claim 5 in the manufacture of a medicament for treating a subject with T cell therapy in need thereof, wherein the medicament is to be administered to the subject..

16. Use of the pharmaceutical composition of claim 8 or 9 in the manufacture of a medicament for treating a subject with T cell therapy in need thereof, wherein the medicament is to be administered to the subject.

17. Use of a therapeutically effective amount of a cell or population of cells in the manufacture of a medicament for treating a subject with T cell therapy in need thereof, wherein the subject is in need of a stimulated immune response, wherein the medicament is to be administered to the subject, wherein the cell is a T cell, wherein a transgene is integrated at a first site within the genome of the T cell such that expression of the transgene is under control of an endogenous promoter of the T cell, wherein the transgene encodes a therapeutic protein or therapeutic nucleic acid.

18. The use of claim 17, wherein the T cell is an immunostimulatory T cell.

19. The use of claim 18, wherein the T cell is selected from the group consisting of cytotoxic T lymphocyte (CTL), CD4⁺ subtype, CD8⁺ subtype, central memory T cell (TCM), stem memory T cell (TSCM), effector memory T cell, effector T cell, Th1 cell, Th2 cell, Th9 cell, Th17 cell, Th22 cell, and Tfh (follicular helper) cell.

20. Use of a therapeutically effective amount of a cell or population of cells in the manufacture of a medicament for treating a subject with T cell therapy in need thereof, wherein the subject is in need of an inhibited immune response, wherein the medicament is to be administered to the subject, wherein the cell is a T cell, wherein a transgene is integrated at a first site within the genome of the T cell such that expression of the transgene is under control of an endogenous promoter of the T cell, wherein the transgene encodes a therapeutic protein or therapeutic nucleic acid.

21. The use of claim 20, wherein the T cell is an immunoinhibitory T cell.

22. The use of claim 21, wherein the T cell is a regulatory T cell.

23. A method of generating a T cell that expresses a therapeutic transgene, comprising:

introducing into a T cell:

(i) a transgene, and

(ii) a homologous recombination system suitable for targeted integration of the transgene at a site within the genome of the cell, whereby the homologous recombination system integrates the transgene at said site within the genome of the cell, and wherein expression of the transgene is under the control of an endogenous promoter, wherein the transgene encodes a therapeutic protein or a therapeutic nucleic acid.

24. A vector comprising a non-integrating gamma-retrovirus.

25. The vector of claim 24, wherein the non-integrating gamma-retrovirus comprises a mutated integrase.

26. A T cell wherein a recombinant nucleic acid sequence encoding a chimeric antigen receptor (CAR) is integrated at a first site within the genome of the cell such that the CAR is expressed by the cell at the surface of the cell, and wherein integration of the nucleic

acid encoding the CAR at said first site reduces or prevents expression of a functional T cell receptor (TCR) complex at the surface of the cell.

27. A human T cell wherein a promotor-less recombinant nucleic acid sequence encoding a CAR is integrated at a site in the genome of the cell, said site being the first exon of the TCR alpha chain, such that the CAR is expressed under control of the endogenous TCR alpha chain promoter, to produce said CAR at the surface of the cell, and wherein integration of the CAR at said site reduces or prevents expression of a functional TCR alpha chain.

28. An isolated population of T cells, which comprises a plurality of the cell of claim 26 or 27.

29. A pharmaceutical composition comprising a therapeutically effective amount of the cell of claim 26 or 27; and a pharmaceutically acceptable carrier.

30. A pharmaceutical composition comprising a therapeutically effective amount of a population of T cells, which population comprises a plurality of the cell of claim 26 or 27; and a pharmaceutically acceptable carrier.

31. Use of an effective amount of the cell of claim 26 or 27 in the manufacture of a medicament for treating a subject with CAR therapy in need thereof, wherein the medicament is to be administered to the subject.

32. Use of the pharmaceutical composition of claim 29 in the manufacture of a medicament for treating a subject with CAR therapy in need thereof, wherein the medicament is to be administered to the subject.

33. Use of a therapeutically effective amount of the cell population of claim 28 in the manufacture of a medicament for treating a subject with CAR therapy in need thereof, wherein the medicament is to be administered to the subject.

34. Use of the pharmaceutical composition of claim 30 in the manufacture of a medicament for treating a subject with CAR therapy in need thereof, wherein the medicament is to be administered to the subject.

35. A method of generating a T cell that expresses a chimeric antigen receptor (CAR) and lacks a functional T cell receptor (TCR) complex, comprising:

introducing into a T cell:

(i) a nucleic acid sequence encoding a CAR, and

(ii) a homologous recombination system suitable for targeted integration of the nucleic acid sequence at a site within the genome of the cell, whereby the homologous recombination system integrates the nucleic acid sequence encoding the CAR at said site within the genome of the cell such that integration of the CAR at said site reduces or prevents expression of a functional T cell receptor complex at the surface of the cell, thereby generating a T cell that expresses the CAR and lacks a functional TCR complex.

36. An induced pluripotent stem cell, wherein a recombinant nucleic acid sequence encoding a chimeric antigen receptor (CAR) is integrated at a first site within the genome of the cell such that the CAR is expressed by the cell at the surface of the cell, and wherein integration of the nucleic acid encoding the CAR at said first site reduces or prevents expression of a functional T cell receptor (TCR) complex at the surface of the cell.