diseases (e.g. EBV, CMV, HIV), bacterial diseases, protozoan diseases, parasitic diseases, fungal diseases, mycoplasma diseases and prion diseases; inflammatory diseases (e.g. chronic inflammatory diseases and acute inflammatory diseases); and autoimmune diseases (e.g. cardiovascular diseases, rheumatoid diseases, glandular diseases, gastrointestinal diseases, cutaneous diseases, hepatic diseases, neurological diseases, muscular diseases, nephric diseases, diseases related to reproduction, connective tissue diseases and systemic diseases).

Thus, the method of the present invention can furthermore be advantageously applied towards treating a disease in a subject while concomitantly facilitating engraftment of a transplant of cells or tissues syngeneic with the anti-third party Tcm cells (e.g. in situations where the cell or tissue transplant and the anti-third party cells are derived from the same donor).

As used herein the term “about” refers to ± 10 %.

The terms "comprises", "comprising", "includes", "including", “having” and their conjugates mean "including but not limited to".

The term “consisting of means “including and limited to”.

~~The term "consisting essentially of" means that the composition, method or structure may include additional ingredients, steps and/or parts, but only if the additional ingredients, steps and/or parts do not materially alter the basic and novel characteristics of the claimed composition, method or structure.~~

~~As used herein, the singular form "a", "an" and "the" include plural references unless the context clearly dictates otherwise. For example, the term "a compound" or "at least one compound" may include a plurality of compounds, including mixtures thereof.~~

Throughout this application, various embodiments of this invention may be presented in a range format. It should be understood that the description in range format is merely for convenience and brevity and should not be construed as an inflexible limitation on the scope of the invention. Accordingly, the description of a range should be considered to have specifically disclosed all the possible subranges as well as individual numerical values within that range. For example, description of a range such as from 1 to 6 should be considered to have specifically disclosed subranges such as from 1 to 3, from 1 to 4, from 1 to 5, from 2 to 4, from 2 to 6, from 3 to 6 etc., as well

clinically available activation markers like CD25 or IFN gamma capture to the depletion step.

Conclusions:

The use of CD137 depletion at the end of Tcm generation might afford a feasible approach to further deplete these cells of alloreactivity.

Attempts to continue refining the use of CD137 depletion in conjunction with CD25 depletion, both of which are available as GMP reagents, are explored.

In addition, experiments are carried out to minimize potential cross reactivity by using artificial APC bearing only one HLA-I allele.

~~Although the invention has been described in conjunction with specific embodiments thereof, it is evident that many alternatives, modifications and variations will be apparent to those skilled in the art. Accordingly, it is intended to embrace all such alternatives, modifications and variations that fall within the spirit and broad scope of the appended claims.~~

~~All publications, patents and patent applications mentioned in this specification are herein incorporated in their entirety by into the specification, to the same extent as if each individual publication, patent or patent application was specifically and individually indicated to be incorporated herein by reference.~~ In addition, citation or identification of any reference in this application shall not be construed as an admission that such reference is available as prior art to the present invention. To the extent that section headings are used, they should not be construed as necessarily limiting.

diseases (e.g. EBV, CMV, HIV), bacterial diseases, protozoan diseases, parasitic diseases, fungal diseases, mycoplasma diseases and prion diseases; inflammatory diseases (e.g. chronic inflammatory diseases and acute inflammatory diseases); and autoimmune diseases (e.g. cardiovascular diseases, rheumatoid diseases, glandular diseases, gastrointestinal diseases, cutaneous diseases, hepatic diseases, neurological diseases, muscular diseases, nephric diseases, diseases related to reproduction, connective tissue diseases and systemic diseases).

Thus, the method of the present invention can furthermore be advantageously applied towards treating a disease in a subject while concomitantly facilitating engraftment of a transplant of cells or tissues syngeneic with the anti-third party Tcm cells (e.g. in situations where the cell or tissue transplant and the anti-third party cells are derived from the same donor).

As used herein the term "about" refers to ± 10 %.

The terms "comprises", "comprising", "includes", "including", "having" and their conjugates mean "including but not limited to".

The term "consisting of means "including and limited to".

The term "consisting essentially of" means that the composition, method or structure may include additional ingredients, steps and/or parts, but only if the additional ingredients, steps and/or parts do not materially alter the basic and novel characteristics of the claimed composition, method or structure.

As used herein, the singular form "a", "an" and "the" include plural references unless the context clearly dictates otherwise. For example, the term "a compound" or "at least one compound" may include a plurality of compounds, including mixtures thereof.

Throughout this application, various embodiments of this invention may be presented in a range format. It should be understood that the description in range format is merely for convenience and brevity and should not be construed as an inflexible limitation on the scope of the invention. Accordingly, the description of a range should be considered to have specifically disclosed all the possible subranges as well as individual numerical values within that range. For example, description of a range such as from 1 to 6 should be considered to have specifically disclosed subranges such as from 1 to 3, from 1 to 4, from 1 to 5, from 2 to 4, from 2 to 6, from 3 to 6 etc., as well

clinically available activation markers like CD25 or IFN gamma capture to the depletion step.

Conclusions:

The use of CD137 depletion at the end of Tcm generation might afford a feasible approach to further deplete these cells of alloreactivity.

Attempts to continue refining the use of CD137 depletion in conjunction with CD25 depletion, both of which are available as GMP reagents, are explored.

In addition, experiments are carried out to minimize potential cross reactivity by using artificial APC bearing only one HLA-I allele.

Although the invention has been described in conjunction with specific embodiments thereof, it is evident that many alternatives, modifications and variations will be apparent to those skilled in the art. Accordingly, it is intended to embrace all such alternatives, modifications and variations that fall within the spirit and broad scope of the appended claims.

All publications, patents and patent applications mentioned in this specification are herein incorporated in their entirety by into the specification, to the same extent as if each individual publication, patent or patent application was specifically and individually indicated to be incorporated herein by reference. In addition, citation or identification of any reference in this application shall not be construed as an admission that such reference is available as prior art to the present invention. To the extent that section headings are used, they should not be construed as necessarily limiting.

diseases (e.g. EBV, CMV, HIV), bacterial diseases, protozoan diseases, parasitic diseases, fungal diseases, mycoplasma diseases and prion diseases; inflammatory diseases (e.g. chronic inflammatory diseases and acute inflammatory diseases); and autoimmune diseases (e.g. cardiovascular diseases, rheumatoid diseases, glandular diseases, gastrointestinal diseases, cutaneous diseases, hepatic diseases, neurological diseases, muscular diseases, nephric diseases, diseases related to reproduction, connective tissue diseases and systemic diseases).

Thus, the method of the present invention can furthermore be advantageously applied towards treating a disease in a subject while concomitantly facilitating engraftment of a transplant of cells or tissues syngeneic with the anti-third party Tcm cells (e.g. in situations where the cell or tissue transplant and the anti-third party cells are derived from the same donor).

As used herein the term “about” refers to ± 10 %.

The terms "comprises", "comprising", "includes", "including", “having” and their conjugates mean "including but not limited to".

The term “consisting of means “including and limited to”.

Throughout this application, various embodiments of this invention may be presented in a range format. It should be understood that the description in range format is merely for convenience and brevity and should not be construed as an inflexible limitation on the scope of the invention. Accordingly, the description of a range should be considered to have specifically disclosed all the possible subranges as well as individual numerical values within that range. For example, description of a range such as from 1 to 6 should be considered to have specifically disclosed subranges such as from 1 to 3, from 1 to 4, from 1 to 5, from 2 to 4, from 2 to 6, from 3 to 6 etc., as well

clinically available activation markers like CD25 or IFN gamma capture to the depletion step.

Conclusions:

The use of CD137 depletion at the end of Tcm generation might afford a feasible approach to further deplete these cells of alloreactivity.

Attempts to continue refining the use of CD137 depletion in conjunction with CD25 depletion, both of which are available as GMP reagents, are explored.

In addition, experiments are carried out to minimize potential cross reactivity by using artificial APC bearing only one HLA-I allele.

In addition, citation or identification of any reference in this application shall not be construed as an admission that such reference is available as prior art to the present invention. To the extent that section headings are used, they should not be construed as necessarily limiting.

WHAT IS CLAIMED IS:

1. A method of generating an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) contacting peripheral blood mononuclear cells (PBMC) with a third party antigen or antigens in the presence of IL-21 so as to allow enrichment of antigen reactive cells; and

(b) culturing said cells resulting from step (a) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

2. The method of claim 1, further comprising depleting non-adherent cells from said PBMC prior to step (a).

3. The method of claim 1 or 2, further comprising depleting CD4+ ~~and/~~or CD56+ cells from said PBMC prior to step (a).

4. The method of claim 1, 2 or 3, further comprising selecting CD45RA+ ~~and/~~or CD45RO- cells from said PBMC prior to step (a).

5. The method of claim 1, wherein said PBMC comprise CD8+ T cells.

6. The method of claim 1, further comprising culturing said cells resulting from step (a) with a third party antigen or antigens in the presence of IL-21, IL-15 and IL-7 following step (a) and prior to step (b).

7. The method of claim 1, wherein said third party antigen or antigens comprise dendritic cells.

8. The method of claim 7, wherein said dendritic cells are irradiated dendritic cells.

9. The method of claim 1, wherein said third party antigen or antigens is selected from the group consisting of third party cells, a cell antigen, a viral antigen, a bacterial antigen, a protein extract, a purified protein and a synthetic peptide presented by autologous presenting cells, non-autologous presenting cells or on an artificial vehicle or on artificial antigen presenting cells.

10. The method of claim 9, wherein said third party cells are stimulatory cells selected from the group consisting of cells purified from peripheral blood lymphocytes, spleen or lymph nodes, cytokine-mobilized PBLs, in vitro expanded antigen-presenting cells (APC), in vitro expanded dendritic cells and artificial antigen presenting cells.

11. A method of generating an isolated population of cells comprising <u>non-GVHD inducing</u> anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with graft-versus-leukemia (GVL) activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ ~~and/~~or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with third party dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said third party dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells

comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

12. A method of generating an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ ~~and/~~or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with non-syngeneic dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said non-syngeneic dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

13. The method of claim 11 or 12, further comprising selecting CD45RA+ ~~and/~~or CD45RO- cells from said PBMC following step (a) and prior to step (b).

14. The method of claim 11 or 12, wherein said CD8+ T cells comprise naïve CD8+ T cells.

15. The method of claim 11 or 12, wherein said dendritic cells comprise in vitro expanded dendritic cells.

16. The method of claim 11 or 12, wherein said dendritic cells comprise irradiated dendritic cells.

17. The method of claim 1, wherein said contacting in said presence of IL-21 is effected for 12 hours to 5 days.

18. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 2-3 days.

19. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 3 days.

20. The method of claim 1, further comprising selecting for activated cells following step (a) and prior to step (b).

21. The method of claim 11, further comprising selecting for activated cells following step (b) and prior to step (c).

22. The method of claim 20 or 21, wherein said selecting for activated cells is effected by selection of CD137+ ~~and/~~or CD25+ cells.

23. The method of claim 20 or 21, wherein said selecting for activated cells is effected 12-72 hours after said contacting.

24. The method of claim 6, wherein said culturing with said third party antigen or antigens in said presence of IL-21, IL-15 and IL-7 is effected for 12 hours to 3 days.

25. The method of claim 1, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 5-20 days.

26. The method of claim 1, 11 or 12, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 7-11 days.

27. The method of claim 1, further comprising depleting alloreactive cells following step (b).

28. The method of claim 11, further comprising depleting alloreactive cells following step (d).

29. The method of claim 27 or 28, wherein said depleting said alloreactive cells is effected by depletion of CD137+ ~~and/~~or CD25+ cells following contacting said cells comprising said central memory T-lymphocyte (Tcm) with host antigen presenting cells (APCs).

30. The method of claim 1 or 12, wherein said peripheral blood mononuclear cells (PBMC) are syngeneic with respect to a subject.

31. The method of claim 1 or 11, wherein said peripheral blood mononuclear cells (PBMC) are non-syngeneic with respect to a subject.

32. The method of claim 11 or 31, wherein said non-syngeneic PBMC are xenogeneic or allogeneic with respect to a subject.

33. The method of claim 1, 11 or 12, wherein said anti-third party cells having a T central memory phenotype comprises a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature.

34. The method of claim 33, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % have said signature.

35. An isolated population of cells comprising <u>non-GVHD inducing</u> anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % comprise a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature, and further wherein said cells

are tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation.

36. An isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, generated according to the method of claims 1-34.

37. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for ~~A method of~~ treating a disease in a subject in need thereof, wherein the disease is selected from the group consisting of a malignant disease, a viral disease and an autoimmune disease~~, the method comprising administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject~~.

38. The ~~method~~ use of claim 37, wherein said malignant disease comprises a leukemia or a lymphoma.

39. The ~~method~~ use of claim 37, wherein said isolated population of cells are syngeneic with the subject.

40. The ~~method~~ use of claim 37, wherein said isolated population of cells are non-syngeneic with the subject.

41. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for ~~A method of~~ treating a subject in need of a cell or tissue transplantation~~, the method comprising:~~

~~(a) transplanting a cell or tissue transplant into the subject; and~~

~~(b) administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject~~.

42. The ~~method~~ use of claim 41, further comprising a ~~conditioning the subject under~~ sublethal, lethal or supralethal ~~conditions~~ conditioning protocol~~prior to said transplanting~~.

43. The ~~method~~ use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject.

44. The ~~method~~ use of claim 41, wherein said cell or tissue transplant is derived from a donor selected from the group consisting of an HLA identical allogeneic donor, an HLA non-identical allogeneic donor and a xenogeneic donor.

45. The ~~method~~ use of claim 41, wherein said cell or tissue transplant comprises immature hematopoietic cells.

46. The ~~method~~ use of claim 41, wherein said cell or tissue transplant is selected from the group consisting of a liver, a pancreas, a spleen, a kidney, a heart, a lung, a skin, an intestine and a lymphoid/hematopoietic tissue or organ.

47. The ~~method~~ use of claim 41, wherein said cell or tissue transplant comprises a co-transplantation of several organs.

48. The ~~method~~ use of claim 47, wherein said co-transplantation comprises transplantation of immature hematopoietic cells and a solid organ.

49. The ~~method~~ use of claim 48, wherein said immature hematopoietic cells and said solid organ or obtained from the same donor.

~~50. The method of claim 48, wherein said immature hematopoietic cells are transplanted prior to, concomitantly with, or following said transplantation of said solid organ.~~

~~51. The method of claim 41, wherein said isolated population of cells are administered prior to, concomitantly with, or following said cell or tissue transplant.~~

~~52~~50. The ~~method~~ use of claim 41, wherein said isolated population of cells are syngeneic with the subject.

~~53~~51. The ~~method~~ use of claim 41, wherein said isolated population of cells are non-syngeneic with the subject.

~~54~~52. The ~~method~~ use of claim 41, wherein said cell or tissue transplant and said isolated population of cells are derived from the same donor.

~~55~~53. The ~~method~~ use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are non-syngeneic with the subject.

~~56~~54. The ~~method~~ use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are syngeneic with the subject.

~~57~~55. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for ~~A method of~~ treating a subject in need of an immature hematopoietic cell transplantation~~, the method comprising:~~

~~(a) transplanting immature hematopoietic cells into the subject; and~~

~~(b) administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject~~.

~~58. The method of claim 57, wherein said isolated population of cells are administered prior to, concomitantly with, or following said immature hematopoietic cells.~~

~~59~~56. The ~~method~~ use of claim ~~57~~55, wherein said immature hematopoietic cells and said isolated population of cells are derived from the same donor.

~~60~~57. The ~~method~~ use of claim ~~59~~56, wherein said donor is non-syngeneic with the subject.

~~61~~58. The ~~method~~ use of claim ~~57~~55, wherein said immature hematopoietic cells and said isolated population of cells are derived from the subject.

~~62~~59. The ~~method~~ use of claim ~~57~~55, further comprising a~~conditioning the subject under~~ sublethal, lethal or supralethal ~~conditions~~ conditioning protocol~~prior to said transplanting~~.

~~63~~60. The ~~method~~ use of claims 37, 41 or ~~57~~55, wherein said subject is a human subject.

WHAT IS CLAIMED IS:

1. A method of generating an isolated population of cells comprising anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) contacting peripheral blood mononuclear cells (PBMC) with a third party antigen or antigens in the presence of IL-21 so as to allow enrichment of antigen reactive cells; and

(b) culturing said cells resulting from step (a) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

2. The method of claim 1, further comprising depleting non-adherent cells from said PBMC prior to step (a).

3. The method of claim 1 or 2, further comprising depleting CD4+ and/or CD56+ cells from said PBMC prior to step (a).

4. The method of claim 1, 2 or 3, further comprising selecting CD45RA+ and/or CD45RO- cells from said PBMC prior to step (a).

5. The method of claim 1, wherein said PBMC comprise CD8+ T cells.

6. The method of claim 1, further comprising culturing said cells resulting from step (a) with a third party antigen or antigens in the presence of IL-21, IL-15 and IL-7 following step (a) and prior to step (b).

7. The method of claim 1, wherein said third party antigen or antigens comprise dendritic cells.

8. The method of claim 7, wherein said dendritic cells are irradiated dendritic cells.

9. The method of claim 1, wherein said third party antigen or antigens is selected from the group consisting of third party cells, a cell antigen, a viral antigen, a bacterial antigen, a protein extract, a purified protein and a synthetic peptide presented by autologous presenting cells, non-autologous presenting cells or on an artificial vehicle or on artificial antigen presenting cells.

10. The method of claim 9, wherein said third party cells are stimulatory cells selected from the group consisting of cells purified from peripheral blood lymphocytes, spleen or lymph nodes, cytokine-mobilized PBLs, in vitro expanded antigen-presenting cells (APC), in vitro expanded dendritic cells and artificial antigen presenting cells.

11. A method of generating an isolated population of cells comprising anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with graft-versus-leukemia (GVL) activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ and/or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with third party dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said third party dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

12. A method of generating an isolated population of cells comprising anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ and/or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with non-syngeneic dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said non-syngeneic dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

13. The method of claim 11 or 12, further comprising selecting CD45RA+ and/or CD45RO- cells from said PBMC following step (a) and prior to step (b).

14. The method of claim 11 or 12, wherein said CD8+ T cells comprise naïve CD8+ T cells.

15. The method of claim 11 or 12, wherein said dendritic cells comprise in vitro expanded dendritic cells.

16. The method of claim 11 or 12, wherein said dendritic cells comprise irradiated dendritic cells.

17. The method of claim 1, wherein said contacting in said presence of IL-21 is effected for 12 hours to 5 days.

18. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 2-3 days.

19. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 3 days.

20. The method of claim 1, further comprising selecting for activated cells following step (a) and prior to step (b).

21. The method of claim 11, further comprising selecting for activated cells following step (b) and prior to step (c).

22. The method of claim 20 or 21, wherein said selecting for activated cells is effected by selection of CD137+ and/or CD25+ cells.

23. The method of claim 20 or 21, wherein said selecting for activated cells is effected 12-72 hours after said contacting.

24. The method of claim 6, wherein said culturing with said third party antigen or antigens in said presence of IL-21, IL-15 and IL-7 is effected for 12 hours to 3 days.

25. The method of claim 1, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 5-20 days.

26. The method of claim 1, 11 or 12, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 7-11 days.

27. The method of claim 1, further comprising depleting alloreactive cells following step (b).

28. The method of claim 11, further comprising depleting alloreactive cells following step (d).

29. The method of claim 27 or 28, wherein said depleting said alloreactive cells is effected by depletion of CD137+ and/or CD25+ cells following contacting said cells comprising said central memory T-lymphocyte (Tcm) with host antigen presenting cells (APCs).

30. The method of claim 1 or 12, wherein said peripheral blood mononuclear cells (PBMC) are syngeneic with respect to a subject.

31. The method of claim 1 or 11, wherein said peripheral blood mononuclear cells (PBMC) are non-syngeneic with respect to a subject.

32. The method of claim 11 or 31, wherein said non-syngeneic PBMC are xenogeneic or allogeneic with respect to a subject.

33. The method of claim 1, 11 or 12, wherein said anti-third party cells having a T central memory phenotype comprises a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature.

34. The method of claim 33, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % have said signature.

35. An isolated population of cells comprising anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % comprise a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature, and further wherein said cells are tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation.

36. An isolated population of cells comprising anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, generated according to the method of claims 1-34.

37. A method of treating a disease in a subject in need thereof, wherein the disease is selected from the group consisting of a malignant disease, a viral disease and an autoimmune disease, the method comprising administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject.

38. The method of claim 37, wherein said malignant disease comprises a leukemia or a lymphoma.

39. The method of claim 37, wherein said isolated population of cells are syngeneic with the subject.

40. The method of claim 37, wherein said isolated population of cells are non-syngeneic with the subject.

41. A method of treating a subject in need of a cell or tissue transplantation, the method comprising:

(a) transplanting a cell or tissue transplant into the subject; and

(b) administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject.

42. The method of claim 41, further comprising conditioning the subject under sublethal, lethal or supralethal conditions prior to said transplanting.

43. The method of claim 41, wherein said cell or tissue transplant is syngeneic with the subject.

44. The method of claim 41, wherein said cell or tissue transplant is derived from a donor selected from the group consisting of an HLA identical allogeneic donor, an HLA non-identical allogeneic donor and a xenogeneic donor.

45. The method of claim 41, wherein said cell or tissue transplant comprises immature hematopoietic cells.

46. The method of claim 41, wherein said cell or tissue transplant is selected from the group consisting of a liver, a pancreas, a spleen, a kidney, a heart, a lung, a skin, an intestine and a lymphoid/hematopoietic tissue or organ.

47. The method of claim 41, wherein said cell or tissue transplant comprises a co-transplantation of several organs.

48. The method of claim 47, wherein said co-transplantation comprises transplantation of immature hematopoietic cells and a solid organ.

49. The method of claim 48, wherein said immature hematopoietic cells and said solid organ or obtained from the same donor.

50. The method of claim 48, wherein said immature hematopoietic cells are transplanted prior to, concomitantly with, or following said transplantation of said solid organ.

51. The method of claim 41, wherein said isolated population of cells are administered prior to, concomitantly with, or following said cell or tissue transplant.

52. The method of claim 41, wherein said isolated population of cells are syngeneic with the subject.

53. The method of claim 41, wherein said isolated population of cells are non-syngeneic with the subject.

54. The method of claim 41, wherein said cell or tissue transplant and said isolated population of cells are derived from the same donor.

55. The method of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are non-syngeneic with the subject.

56. The method of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are syngeneic with the subject.

57. A method of treating a subject in need of an immature hematopoietic cell transplantation, the method comprising:

(a) transplanting immature hematopoietic cells into the subject; and

(b) administering to the subject a therapeutically effective amount of the isolated population of cells of claim 35 or 36, thereby treating the subject.

58. The method of claim 57, wherein said isolated population of cells are administered prior to, concomitantly with, or following said immature hematopoietic cells.

59. The method of claim 57, wherein said immature hematopoietic cells and said isolated population of cells are derived from the same donor.

60. The method of claim 59, wherein said donor is non-syngeneic with the subject.

61. The method of claim 57, wherein said immature hematopoietic cells and said isolated population of cells are derived from the subject.

62. The method of claim 57, further comprising conditioning the subject under sublethal, lethal or supralethal conditions prior to said transplanting.

63. The method of claims 37, 41 or 57, wherein said subject is a human subject.

WHAT IS CLAIMED IS:

1. A method of generating an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) contacting peripheral blood mononuclear cells (PBMC) with a third party antigen or antigens in the presence of IL-21 so as to allow enrichment of antigen reactive cells; and

(b) culturing said cells resulting from step (a) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

2. The method of claim 1, further comprising depleting non-adherent cells from said PBMC prior to step (a).

3. The method of claim 1 or 2, further comprising depleting CD4+ or CD56+ cells from said PBMC prior to step (a).

4. The method of claim 1, 2 or 3, further comprising selecting CD45RA+ or CD45RO- cells from said PBMC prior to step (a).

5. The method of claim 1, wherein said PBMC comprise CD8+ T cells.

6. The method of claim 1, further comprising culturing said cells resulting from step (a) with a third party antigen or antigens in the presence of IL-21, IL-15 and IL-7 following step (a) and prior to step (b).

7. The method of claim 1, wherein said third party antigen or antigens comprise dendritic cells.

8. The method of claim 7, wherein said dendritic cells are irradiated dendritic cells.

9. The method of claim 1, wherein said third party antigen or antigens is selected from the group consisting of third party cells, a cell antigen, a viral antigen, a bacterial antigen, a protein extract, a purified protein and a synthetic peptide presented by autologous presenting cells, non-autologous presenting cells or on an artificial vehicle or on artificial antigen presenting cells.

10. The method of claim 9, wherein said third party cells are stimulatory cells selected from the group consisting of cells purified from peripheral blood lymphocytes, spleen or lymph nodes, cytokine-mobilized PBLs, in vitro expanded antigen-presenting cells (APC), in vitro expanded dendritic cells and artificial antigen presenting cells.

11. A method of generating an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with graft-versus-leukemia (GVL) activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with third party dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said third party dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells

comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

12. A method of generating an isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, the method comprising:

(a) treating non-adherent peripheral blood mononuclear cells (PBMC) with an agent capable of depleting CD4+ or CD56+ cells so as to obtain CD8+ T cells;

(b) contacting said CD8+ T cells with non-syngeneic dendritic cells in the presence of IL-21 for 12 hours to 5 days so as to allow enrichment of antigen reactive cells;

(c) culturing said cells resulting from step (b) with said non-syngeneic dendritic cells in the presence of IL-21, IL-15 and IL-7 for 12 hours to 3 days; and

(d) culturing said cells resulting from step (c) in the presence of IL-21, IL-15 and IL-7 in an antigen free environment for 5-20 days so as to allow proliferation of cells comprising said central memory T-lymphocyte (Tcm) phenotype, thereby generating the isolated population of cells.

13. The method of claim 11 or 12, further comprising selecting CD45RA+ or CD45RO- cells from said PBMC following step (a) and prior to step (b).

14. The method of claim 11 or 12, wherein said CD8+ T cells comprise naïve CD8+ T cells.

15. The method of claim 11 or 12, wherein said dendritic cells comprise in vitro expanded dendritic cells.

16. The method of claim 11 or 12, wherein said dendritic cells comprise irradiated dendritic cells.

17. The method of claim 1, wherein said contacting in said presence of IL-21 is effected for 12 hours to 5 days.

18. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 2-3 days.

19. The method of claim 1, 11 or 12, wherein said contacting in said presence of IL-21 is effected for 3 days.

20. The method of claim 1, further comprising selecting for activated cells following step (a) and prior to step (b).

21. The method of claim 11, further comprising selecting for activated cells following step (b) and prior to step (c).

22. The method of claim 20 or 21, wherein said selecting for activated cells is effected by selection of CD137+ or CD25+ cells.

23. The method of claim 20 or 21, wherein said selecting for activated cells is effected 12-72 hours after said contacting.

24. The method of claim 6, wherein said culturing with said third party antigen or antigens in said presence of IL-21, IL-15 and IL-7 is effected for 12 hours to 3 days.

25. The method of claim 1, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 5-20 days.

26. The method of claim 1, 11 or 12, wherein said culturing in said presence of IL-21, IL-15 and IL-7 in said antigen free environment is effected for 7-11 days.

27. The method of claim 1, further comprising depleting alloreactive cells following step (b).

28. The method of claim 11, further comprising depleting alloreactive cells following step (d).

29. The method of claim 27 or 28, wherein said depleting said alloreactive cells is effected by depletion of CD137+ or CD25+ cells following contacting said cells comprising said central memory T-lymphocyte (Tcm) with host antigen presenting cells (APCs).

30. The method of claim 1 or 12, wherein said peripheral blood mononuclear cells (PBMC) are syngeneic with respect to a subject.

31. The method of claim 1 or 11, wherein said peripheral blood mononuclear cells (PBMC) are non-syngeneic with respect to a subject.

32. The method of claim 11 or 31, wherein said non-syngeneic PBMC are xenogeneic or allogeneic with respect to a subject.

33. The method of claim 1, 11 or 12, wherein said anti-third party cells having a T central memory phenotype comprises a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature.

34. The method of claim 33, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % have said signature.

35. An isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, wherein at least 50 % of the isolated population of cells are CD3+CD8+ cells of which at least 50 % comprise a $CD3^+$, $CD8^+$, $CD62L^+$, $CD45RA^-$, $CD45RO^+$ signature, and further wherein said cells

are tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation.

36. An isolated population of cells comprising non-GVHD inducing anti-third party cells having a central memory T-lymphocyte (Tcm) phenotype, said cells being tolerance-inducing cells and/or endowed with anti-disease activity, and capable of homing to the lymph nodes following transplantation, generated according to the method of claims 1-34.

37. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for treating a disease in a subject in need thereof, wherein the disease is selected from the group consisting of a malignant disease, a viral disease and an autoimmune disease.

38. The use of claim 37, wherein said malignant disease comprises a leukemia or a lymphoma.

39. The use of claim 37, wherein said isolated population of cells are syngeneic with the subject.

40. The use of claim 37, wherein said isolated population of cells are non-syngeneic with the subject.

41. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for treating a subject in need of a cell or tissue transplantation.

42. The use of claim 41, further comprising a sublethal, lethal or supralethal conditioning protocol.

43. The use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject.

44. The use of claim 41, wherein said cell or tissue transplant is derived from a donor selected from the group consisting of an HLA identical allogeneic donor, an HLA non-identical allogeneic donor and a xenogeneic donor.

45. The use of claim 41, wherein said cell or tissue transplant comprises immature hematopoietic cells.

46. The use of claim 41, wherein said cell or tissue transplant is selected from the group consisting of a liver, a pancreas, a spleen, a kidney, a heart, a lung, a skin, an intestine and a lymphoid/hematopoietic tissue or organ.

47. The use of claim 41, wherein said cell or tissue transplant comprises a co-transplantation of several organs.

48. The use of claim 47, wherein said co-transplantation comprises transplantation of immature hematopoietic cells and a solid organ.

49. The use of claim 48, wherein said immature hematopoietic cells and said solid organ or obtained from the same donor.

50. The use of claim 41, wherein said isolated population of cells are syngeneic with the subject.

51. The use of claim 41, wherein said isolated population of cells are non-syngeneic with the subject.

52. The use of claim 41, wherein said cell or tissue transplant and said isolated population of cells are derived from the same donor.

53. The use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are non-syngeneic with the subject.

54. The use of claim 41, wherein said cell or tissue transplant is syngeneic with the subject and said isolated population of cells are syngeneic with the subject.

55. Use of a therapeutically effective amount of the isolated population of cells of claim 35 or 36 for the manufacture of a medicament identified for treating a subject in need of an immature hematopoietic cell transplantation.

56. The use of claim 55, wherein said immature hematopoietic cells and said isolated population of cells are derived from the same donor.

57. The use of claim 56, wherein said donor is non-syngeneic with the subject.

58. The use of claim 55, wherein said immature hematopoietic cells and said isolated population of cells are derived from the subject.

59. The use of claim 55, further comprising a sublethal, lethal or supralethal conditioning protocol.

60. The use of claims 37, 41 or 55, wherein said subject is a human subject.