CLAIMS

1. A method for the isolation of γδ T cells from a non-haematopoietic tissue sample comprising the steps of:

(i) culturing the non-haematopoietic tissue sample in the presence of Interleukin 1β (IL-1β), Interleukin 2 (IL-2), and Interleukin-15 (IL-15); and

(ii) collecting a population of γδ T cells cultured from the non-haematopoietic tissue sample;

wherein the non-haematopoietic tissue sample is skin, gut or gastrointestinal tract; and

wherein the non-haematopoietic tissue sample is cultured in media containing serum or plasma.

2. The method according to any preceding claim, wherein step (i) further comprises culturing the non-haematopoietic tissue sample in the presence of Interleukin-4 (IL-4) and/or Interferon-γ (IFN-γ).

3. The method according to any preceding claim, wherein the culturing is performed in the absence of Interleukin-21 (IL-21).

4. The method according to claim 1 or claim 2, wherein the culturing is performed in the presence Interleukin-21 (IL-21) at a concentration between 15 ng/mL and 25 ng/mL.

5. The method according to any preceding claim, wherein the non-haematopoietic tissue is cultured in media containing 2.5% human plasma.

6. The method according to any one of claims 1 to 4, wherein the non-haematopoietic tissue is cultured in media containing 10% human AB serum.

7. The method according to any preceding claim, wherein the γδ T cells are collected after at least 7 days of culturing.

8. The method according to any preceding claim, wherein the γδ T cells are collected after at least 14 days of culturing.

9. The method according to any preceding claim, wherein the γδ T cells are collected prior to 35 days of culturing.

10. The method according to any preceding claim, wherein the γδ T cells are collected after 19 days of culturing.

11. The method according to any preceding claim, wherein the γδ T cells are collected after 21 days of culturing.

12. The method according to any preceding claim, wherein the non-haematopoietic tissue sample is an intact biopsy.

13. The method according to any preceding claim, wherein the non-haematopoietic tissue sample is skin.

14. The method according to claim 13, wherein the skin sample comprises the epidermal and dermal layer.

15. The method according to any preceding claim, wherein the non-haematopoietic tissue sample is gut or gastrointestinal tract.

16. The method according to any preceding claim, wherein the non-haematopoietic tissue sample has been obtained from a human.

17. The method according to any preceding claim, wherein the method is performed in a vessel comprising a gas permeable material.

18. The method according to any preceding claim, wherein the isolated population of γδ T cells comprise a population of Vδ1 T cells.

19. The method according to claim 18, wherein less than 80% of the isolated population of Vδ1 T cells expresses CD45RA.

20. The method according to claim 18 or claim 19, wherein less than 10% of the isolated population of Vδ1 T cells expresses NKG2A.

21. The method according to any preceding claim, further comprising expanding the isolated population of γδ T cells.

22. A method for the isolation and expansion of γδ T cells from a non-haematopoietic tissue sample comprising the steps of:

(i) isolating a population of γδ T cells from the non-haematopoietic tissue sample according to the method of any preceding claim; and

(ii) further culturing said population of γδ T cells for at least 5 days to produce an expanded population of γδ T cells.

23. The method according to claim 22, wherein the isolated r γδ T cell population is frozen and then thawed prior to step (ii).

24. The method according to claim 22 or claim 23, wherein the expansion step comprises culturing the γδ T cells in the presence of:

(a) IL-2;

(b) IL-15; and

(c) IL-21

for at least 5 days in amounts effective to produce an expanded population of γδ T cells.

25. The method according to claim 24, which further comprises culturing the γδ T cells in the presence of IL-4.

26. The method according to any one of claims 22 to 25, wherein the expansion step comprises culturing the γδ T cells in the absence of substantial stromal cell contact.

27. The method according to any one of claims 22 to 26, wherein the expansion step comprises the absence of exogenous TCR pathway agonists.

28. A method of isolation for γδ T cells from a non-haematopoietic tissue sample comprising the steps of:

(i) isolating a population of γδ T cells from the non-haematopoietic tissue sample according to the method of any preceding claim; and

(ii) freezing the isolated population of γδ T cells.

29. The method according to claim 28, wherein γδ T cells are isolated by culturing the non-haematopoietic tissue sample in the presence of IL-2 and IL-15.

30. The method according to claim 29, wherein γδ T cells are isolated by culturing the non-haematopoietic tissue sample in the presence of IL-21 at a concentration between 15 ng/mL and 25 ng/mL.

31. The method according to claim 28 to 30, wherein the γδ T cells are frozen after 19 days of isolation.

32. The method according to any one of claims 28 to 30, wherein the γδ T cells are frozen after 21 days of isolation.

33. The method according to any one of claims 28 to 32, wherein the isolated population of γδ T cells are frozen in a freezing solution and stored in liquid nitrogen (vapour phase).

34. The method according to any one of claims 28 to 33, which additionally comprises: (iii) thawing the frozen population of γδ T cells; and (iv) culturing the thawed population of γδ T cells for at least 5 days to produce an expanded population of γδ T cells.