

CLAIMS

1. An isolated antibody that specifically binds to human glucocorticoid-induced TNFR-related protein (GITR), comprising:

(A) (a) a heavy chain variable region (VH) complementarity determining region (CDR) 1 comprising the amino acid sequence of $X_1YX_2MX_3$ (SEQ ID NO: 1), wherein

X_1 is D, E, G or A;

X_2 is A, V, L, I, P, F, M or Y; and

X_3 is Y, G, N, Q, S, T, C, W, F or H;

(b) a VH CDR2 comprising the amino acid sequence of $X_1IX_2X_3X_4SGX_5X_6X_7YX_8QKFX_9X_{10}$ (SEQ ID NO: 2), wherein

X_1 is V, A, L, I, P, F, M or T;

X_2 is R, K, H, Q or A;

X_3 is T, G, N, Q, S, C, W, Y, V, I or P;

X_4 is Y, G, N, Q, S, T, C, W, F, H, or A;

X_5 is D, E, G or A;

X_6 is V, A, L, I, P, F, M or T;

X_7 is T, G, N, Q, S, C, W, Y, V, I, P or A;

X_8 is N, G, Q, S, T, C, W, Y or A;

X_9 is K, R, H, Q or A; and

X_{10} is D, E, G or A;

(c) a VH CDR3 comprising the amino acid sequence of $SGTVRGX_1X_2X_3$ (SEQ ID NO: 3), wherein

X_1 is F, A, V, L, I, P, M, Y, W, H or S;

X_2 is A, or D; and

X_3 is Y, G, N, Q, S, T, C, W, F, H or V;

(d) a light chain variable region (VL) CDR1 comprising the amino acid sequence of $KSSQSX_1X_2X_3X_4X_5X_6X_7KX_8YX_9$ (SEQ ID NO: 4), wherein:

X_1 is L, A, V, I, P, F or M;

X_2 is L, A, V, I, P, F, M or S;

X_3 is N, G, Q, S, T, C, W, Y or A;

X_4 is S, G, N, Q, T, C, W, Y or A;

X₅ is G, N, Q, S, T, C, W, Y or A;

X₆ is N, G, Q, S, T, C, W, Y or A;

X₇ is Q, G, N, S, T, C, W, Y or A;

X₈ is N, G, Q, S, T, C, W, Y or A; and

X₉ is T, G, N, Q, S, C, W, Y, V, I or A;

(e) a VL CDR2 comprising the amino acid sequence of X₁ASTRX₂X₃ (SEQ ID NO: 5), wherein:

X₁ is W, G, N, Q, S, T, C, Y, F, H or A;

X₂ is E, D or A; and

X₃ is S, G, N, Q, T, C, W, Y or A; and

(f) a VL CDR3 comprising the amino acid sequence of QX₁X₂YX₃X₄PYT (SEQ ID NO: 6), wherein:

X₁ is N, G, Q, S, T, C, W or Y;

X₂ is D, E or Y; and

X₃ is S, G, N, Q, T, C, W, Y or A; and

X₄ is Y, G, N, Q, S, T, C, W, F, H, L, or A; or

(B) (a) a VH CDR1 comprising the amino acid sequence of X₁YX₂MX₃ (SEQ ID NO: 7), wherein

X₁ is D, E or G;

X₂ is A or V; and

X₃ is Y or H;

(b) a VH CDR2 comprising the amino acid sequence of X₁IX₂TX₃SGX₄X₅X₆YNQKFX₇X₈ (SEQ ID NO: 8), wherein

X₁ is V or L;

X₂ is R, K or Q;

X₃ is Y or F;

X₄ is D, E or G;

X₅ is V or L;

X₆ is T or S;

X₇ is K, R or Q; and

X₈ is D, E or G;

- (c) a VH CDR3 comprising the amino acid sequence of SGTVRGFAY (SEQ ID NO: 9);
- (d) a VL CDR1 comprising the amino acid sequence of KSSQSLLNSX₁NQKNYLX₂ (SEQ ID NO: 10), wherein
 - X₁ is G or S; and
 - X₂ is T or S;
- (e) a VL CDR2 comprising the amino acid sequence of WASTRES (SEQ ID NO: 11); and
- (f) a VL CDR3 comprising the amino acid sequence of QNX₁YSX₂PYT (SEQ ID NO: 12), wherein
 - X₁ is D or E; and
 - X₂ is Y, F or S.

2. The antibody of claim 1, wherein:

- (a) the VH CDR1 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 13, 19-23, 35, and 116-119;
- (b) the VH CDR2 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 14, 24-33, 114, 115, 120-188, and 194;
- (c) the VH CDR3 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 15, 34, and 189;
- (d) the VL CDR1 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 16 and 101-104;
- (e) the VL CDR2 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 17 and 105; or
- (f) the VL CDR3 comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 18, 106-109, 192, and 193.

3. The antibody of claim 1, comprising a VH comprising:

- the VH CDR1, comprising the amino acid sequence of DYAMY (SEQ ID NO: 13),
- the VH CDR2, comprising the amino acid sequence of VIRTYS GDVTYNQKFKD (SEQ ID NO: 14), and

- the VH CDR3, comprising the amino acid sequence of SGTVRGFAY (SEQ ID NO: 15).
4. The antibody of claim 1, comprising a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 201, 206, and 215-389.
 5. The antibody of claim 4, wherein the VH comprises the amino acid sequence of SEQ ID NO: 206.
 6. The antibody of claim 1, comprising a VL comprising:
 - the VL CDR1, comprising the amino acid sequence of KSSQSLLNSGNQKNYLT (SEQ ID NO: 16),
 - the VL CDR2, comprising the amino acid sequence of WASTRES (SEQ ID NO: 17), and
 - the VL CDR3, comprising the amino acid sequence of QNDYSYPYT (SEQ ID NO: 18).
 7. The antibody of claim 1 or 2, comprising a VL comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 202, 207, 208 and 400-519.
 8. The antibody of claim 7, wherein the VL comprises the amino acid sequence of SEQ ID NO: 208.
 9. The antibody of claim 1,
 - wherein the VH CDR1, VH CDR2, and VH CDR3 each comprises the respective amino acid sequence of the VH CDR1, VH CDR2, and VH CDR3 of an antibody in Table 2;
 - wherein the VH CDR1, VH CDR2, and VH CDR3 each comprises the respective amino acid sequence of the VH CDR1, VH CDR2, and VH CDR3 of an antibody in Table 6;
 - wherein the VL CDR1, VL CDR2, and VL CDR3 each comprises the respective amino acid sequence of the VL CDR1, VL CDR2, and VL CDR3 of an antibody in Table 1;
 - or
 - wherein the VL CDR1, VL CDR2, and VL CDR3 each comprises the respective amino acid sequence of the VL CDR1, VL CDR2, and VL CDR3 of an antibody in Table 5.

10. The antibody of claim 1, comprising:

(a) a VH comprising:

the VH CDR1, comprising the amino acid sequence of DYAMY (SEQ ID NO: 13),

the VH CDR2, comprising the amino acid sequence of VIRTYSGDVTYNQKFKD (SEQ ID NO: 14), and

the VH CDR3, comprising the amino acid sequence of SGTVRGFAY (SEQ ID NO: 15); and

(b) a VL comprising:

the VL CDR1, comprising the amino acid sequence of KSSQSLNLSGNQKNYLT (SEQ ID NO: 16),

the VL CDR2, comprising the amino acid sequence of WASTRES (SEQ ID NO: 17), and

the VL CDR3, comprising the amino acid sequence of QNDYSYPYT (SEQ ID NO: 18).

11. The antibody of claim 10, wherein the VH comprises an amino acid sequence having at least 98% sequence identity to the amino acid sequence of SEQ ID NO: 206.

12. The antibody of claim 11, wherein the VH comprises the amino acid sequence of SEQ ID NO: 206.

13. The antibody of claim 12, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 567.

14. The antibody of claim 12, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 554.

15. The antibody of claim 10, wherein the VL comprises an amino acid sequence having at least 98% sequence identity to the amino acid sequence of SEQ ID NO: 208.

16. The antibody of claim 15, wherein the VL comprises the amino acid sequence of SEQ ID NO: 208.

17. The antibody of claim 16, comprising a light chain comprising the amino acid sequence of SEQ ID NO: 576.

18. The antibody of claim 1, comprising a VH comprising an amino acid sequence having at least 98% sequence identity to the amino acid sequence of SEQ ID NO: 206, and further comprising a VL comprising an amino acid sequence having at least 98% sequence identity to the amino acid sequence of SEQ ID NO: 208.

19. The antibody of claim 18, wherein the VH comprises the amino acid sequence of SEQ ID NO: 206.

20. The antibody of claim 19, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 567.

21. The antibody of claim 19, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 554.

22. The antibody of claim 18, wherein the VL comprises the amino acid sequence of SEQ ID NO: 208.

23. The antibody of claim 22, comprising a light chain comprising the amino acid sequence of SEQ ID NO: 576.

24. The antibody of claim 1, comprising:

- (a) a VH comprising the amino acid sequence of SEQ ID NO: 203; and/or
- (b) a VL comprising the amino acid sequence of SEQ ID NO: 204 or 205.

25. The antibody of claim 24, comprising a VH comprising the amino acid sequence of SEQ ID NO: 206, optionally wherein the antibody comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 554 or 567.

26. The antibody of claim 24, comprising a VL comprising the amino acid sequence of SEQ ID NO: 208, optionally wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 576.

27. The antibody of claim 1, comprising:

(a) a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 201, 206 and 215-389, and/or

(b) a VL comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 202, 207, 208 and 400-519.

28. The antibody of claim 27, wherein the VH comprises the amino acid sequence of SEQ ID NO: 206, optionally wherein the antibody comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 554 or 567.

29. The antibody of claim 27, wherein the VL comprises the amino acid sequence of SEQ ID NO: 208, optionally wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 576.

30. The antibody of claim 1, comprising:

(a) a VH comprising the amino acid sequence of SEQ ID NO: 206; and

(b) a VL comprising the amino acid sequence of SEQ ID NO: 208.

31. The antibody of claim 30, wherein:

(a) the amino acid sequence of the VH consists of the amino acid sequence of SEQ ID NO: 206; and

(b) the amino acid sequence of the VL consists of the amino acid sequence of SEQ ID NO: 208.

32. The antibody of claim 30, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 567.

33. The antibody of claim 30, comprising a heavy chain comprising the amino acid sequence of SEQ ID NO: 554.

34. The antibody of claim 30, comprising a light chain comprising the amino acid sequence of SEQ ID NO: 576.

35. The antibody of claim 1, comprising:

- (a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 567; and
- (b) a light chain comprising the amino acid sequence of SEQ ID NO: 576.

36. The antibody of claim 35, wherein:

- (a) the amino acid sequence of the heavy chain consists of the amino acid sequence of SEQ ID NO: 567; and
- (b) the amino acid sequence of the light chain consists of the amino acid sequence of SEQ ID NO: 576.

37. The antibody of claim 1, comprising:

- (a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 554; and
- (b) a light chain comprising the amino acid sequence of SEQ ID NO: 576.

38. The antibody of claim 37, wherein:

- (a) the amino acid sequence of the heavy chain consists of the amino acid sequence of SEQ ID NO: 554; and
- (b) the amino acid sequence of the light chain consists of the amino acid sequence of SEQ ID NO: 576.

39. The antibody of claim 1, wherein the VH and VL amino acid sequences comprise the amino acid sequences listed in a single row of Figure 23, a single row of any one of Figures 24A-24C, or a single row of Table 17.

40. The antibody of claim 1, wherein:

- (A) the VH CDR1 comprises the amino acid sequence of SEQ ID NO: 20, the VH CDR2 comprises the amino acid sequence of SEQ ID NO: 27, and the VH CDR3 comprises the amino acid sequence of SEQ ID NO: 34, and
the VL CDR1 comprises the amino acid sequence of SEQ ID NO: 103, the VL CDR2 comprises the amino acid sequence of SEQ ID NO: 105, and the VL CDR3 comprises the amino acid sequence of SEQ ID NO: 108;
- (B) the VH CDR1 comprises the amino acid sequence of SEQ ID NO: 22, the VH CDR2 comprises the amino acid sequence of SEQ ID NO: 29, and the VH CDR3 comprises the amino acid sequence of SEQ ID NO: 34, and
the VL CDR1 comprises the amino acid sequence of SEQ ID NO: 102, the VL CDR2 comprises the amino acid sequence of SEQ ID NO: 105, and the VL CDR3 comprises the amino acid sequence of SEQ ID NO: 107; or
- (C) the VH CDR1 comprising the amino acid sequence of SEQ ID NO: 22, the VH CDR2 comprises the amino acid sequence of SEQ ID NO: 33, and the VH CDR3 comprises the amino acid sequence of SEQ ID NO: 34, and
the VL CDR1 comprises the amino acid sequence of SEQ ID NO: 102, the VL CDR2 comprises the amino acid sequence of SEQ ID NO: 105, and the VL CDR3 comprises the amino acid sequence of SEQ ID NO: 107.

41. The antibody of claim 1, comprising:

- (a) a VH comprising the amino acid sequence of SEQ ID NO: 255 and a VL comprising the amino acid sequence of SEQ ID NO: 441;
- (b) a VH comprising the amino acid sequence of SEQ ID NO: 276 and a VL comprising the amino acid sequence of SEQ ID NO: 440; or
- (c) a VH comprising the amino acid sequence of SEQ ID NO: 345 and a VL comprising the amino acid sequence of SEQ ID NO: 440.

42. The antibody of claim 1, comprising a heavy chain comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 553, 568-570, 581, and 582; and/or comprising a light chain comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 555, 556, and 571-575.

43. The antibody of claim 1, comprising:

- (a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 553, and a light chain comprising the amino acid sequence of SEQ ID NO: 556;
- (b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 554, and a light chain comprising the amino acid sequence of SEQ ID NO: 556;
- (c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 553, and a light chain comprising the amino acid sequence of SEQ ID NO: 555;
- (d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 554, and a light chain comprising the amino acid sequence of SEQ ID NO: 555;
- (e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 567, and a light chain comprising the amino acid sequence of SEQ ID NO: 573;
- (f) a heavy chain comprising the amino acid sequence of SEQ ID NO: 581 and a light chain comprising the amino acid sequence of SEQ ID NO: 556;
- (g) a heavy chain comprising the amino acid sequence of SEQ ID NO: 582 and a light chain comprising the amino acid sequence of SEQ ID NO: 556;
- (h) a heavy chain comprising the amino acid sequence of SEQ ID NO: 581 and a light chain comprising the amino acid sequence of SEQ ID NO: 576;
- (i) a heavy chain comprising the amino acid sequence of SEQ ID NO: 582 and a light chain comprising the amino acid sequence of SEQ ID NO: 576; or
- (j) a heavy chain comprising the amino acid sequence of SEQ ID NO: 567 and a light chain comprising the amino acid sequence of SEQ ID NO: 587.

44. The antibody of claim 1, comprising:

- (a) a VH having human derived framework regions;

- (b) a VH comprising the framework regions of a VH comprising the amino acid sequence of SEQ ID NO: 203;
- (c) a VH comprising a framework region that is at least 75%, 80%, 85%, 90%, 95%, or 100% identical to a framework region of a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 201, SEQ ID NO: 206, and SEQ ID NOS: 215-389; or
- (d) a VH comprising a framework region that is or is derived from an amino acid sequence encoded by a human gene, wherein said amino acid sequence is selected from the group consisting of IGHV1-2*02 (SEQ ID NO: 601), IGHV1-3*01 (SEQ ID NO: 602), IGHV1-46*01 (SEQ ID NO: 603), IGHV1-18*01 (SEQ ID NO: 604), IGHV1-69*01 (SEQ ID NO: 605), and IGHV7-4-1*02 (SEQ ID NO: 606);
optionally wherein the antibody further comprises a VH framework region that is derived from the amino acid sequence of SEQ ID NO: 601 and wherein at least one amino acid in the amino acid sequence of SEQ ID NO: 601 is substituted with an amino acid in an analogous position in a corresponding non-human VH framework region;
optionally wherein the amino acid substitution is at an amino acid position selected from the group consisting of 24, 48, 67, 71, 73, and 94, and wherein the amino acid position of each group member is indicated according to the Kabat numbering;
optionally wherein the amino acid substitution is selected from the group consisting of 24G, 48I, 67A, 71V, 73K, and 94K, and wherein the amino acid position of each group member is indicated according to the Kabat numbering.

45. The antibody of claim 1, comprising:

- (a) a VL having human derived framework regions;
- (b) a VL comprising the framework regions of a VL comprising the amino acid sequence of SEQ ID NO: 204 or SEQ ID NO: 205;
- (c) a VL comprising a framework region which is at least 75%, 80%, 85%, 90%, 95%, or 100% identical to a framework region of a VL comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 202, SEQ ID NO: 207, SEQ ID NO: 208, SEQ ID NOS: 400-518, and SEQ ID NO: 519; or

(d) a VL comprising a framework region that is or is derived from an amino acid sequence encoded by a human gene, wherein said amino acid sequence is selected from the group consisting of IGKV4-1*01 (SEQ ID NO: 607) and IGKV3-7*02 (SEQ ID NO: 608);

optionally wherein the antibody comprises a VL framework region that is derived from an amino acid sequence selected from the group consisting of IGKV4-1*01 (SEQ ID NO: 607) and IGKV3-7*02 (SEQ ID NO: 608) and at least one amino acid in the amino acid sequence of SEQ ID NO: 607 or SEQ ID NO: 608 is substituted with an amino acid in an analogous position in a corresponding non-human VL framework region;

optionally wherein the amino acid substitution is at amino acid position 87, and wherein the amino acid position is indicated according to the Kabat numbering;

optionally wherein the amino acid substitution is 87H, and wherein the amino acid position is indicated according to the Kabat numbering.

46. The antibody of claim 1, comprising a heavy chain constant region and/or a light chain constant region;

optionally wherein the heavy chain constant region is selected from the group of human immunoglobulins consisting of IgG₁, IgG₂, IgG₃, IgG₄, IgA₁, and IgA₂;

optionally wherein the IgG₁ is non-fucosylated IgG₁,

optionally wherein the amino acid sequence of IgG₁ comprises a N297A mutation,

optionally wherein the amino acid sequence of IgG₁ comprises a N297Q mutation,

optionally wherein the amino acid sequence of IgG₄ comprises a S228P mutation, or

optionally wherein the amino acid sequence of IgG₂ comprises a C127S mutation;

optionally wherein the heavy chain constant region comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 557-562, 583, and 584; and/or

optionally wherein the light chain constant region is selected from the group of human immunoglobulins consisting of IgGκ and IgGλ.

47. The antibody of claim 46, wherein the heavy chain constant region is an IgG₁ heavy chain constant region.

48. The antibody of claim 1, wherein the antibody is a humanized antibody, murine antibody, or chimeric antibody.

49. An isolated antibody that specifically binds to human GITR, comprising:

- (a) a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 201, 203, 206 and 215-389; or
- (b) a VL comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 202, 204, 205, 207, 208, and 400-519.

50. An isolated antibody that specifically binds to human GITR, comprising a VH comprising the amino acid sequence of SEQ ID NO: 206, or a VL comprising the amino acid sequence of SEQ ID NO: 208.

51. An isolated antibody that specifically binds to human GITR, comprising the VL CDR1, VL CDR2, VL CDR3, VH CDR1, VH CDR2, and VH CDR3 of antibody 22 as set forth in Table 1 and Table 2.

52. An isolated antibody that specifically binds to human GITR, comprising:

- (a) a VH comprising the amino acid sequence of SEQ ID NO: 206; and
- (b) a VL comprising the amino acid sequence of SEQ ID NO: 207.

53. An isolated antibody that specifically binds to human GITR, comprising:

- (a) a heavy chain variable region (VH) comprising a VH CDR1, a VH CDR2, a VH CDR3 and framework regions derived from a human immunoglobulin, wherein the VH CDR1 comprises the amino acid sequence of DYAMY (SEQ ID NO: 13), the VH CDR2 comprises the amino acid sequence of VIRTYSGDVTYNQKFKD (SEQ ID NO: 14) and the VH CDR3 comprises the amino acid sequence of SGTVRGFAY (SEQ ID NO: 15); or
- (b) a light chain variable region (VL) comprising a VL CDR1, a VL CDR2, a VL CDR3 and framework regions derived from a human immunoglobulin, wherein the VL CDR1 comprises the amino acid sequence of KSSQSLNLSGNQKNYLT (SEQ ID NO: 16), the

VL CDR2 comprises the amino acid sequence of WASTRES (SEQ ID NO: 17) and the
VL CDR3 comprises the amino acid sequence of QNDYSYPYT (SEQ ID NO: 18).

54. The antibody of any one of claims 1-53, wherein the antibody is agonistic.

55. The antibody of any one of claims 1-54, wherein:

(a) the antibody induces, activates, or enhances an activity of human GITR;

(b) the antibody induces, activates or enhances an activity of human GITR in a cell independent of TCR triggering;

optionally wherein the cell is a T cell or wherein the cell is not a T cell;

optionally wherein the cell is selected from the group consisting of a B cell, a plasma cell, a memory cell, a natural killer cell, a granulocyte, a neutrophil, an eosinophil, a basophil, a mast cell, a monocyte, a dendritic cell, a plasmacytoid dendritic cell, an NKT cell, and a macrophage;

(c) the antibody induces, activates or enhances an activity of NF- κ B in a cell independent of TCR triggering;

optionally wherein the cell is a T cell or wherein the cell is not a T cell;

optionally wherein the cell is selected from the group consisting of a B cell, a plasma cell, a memory cell, a natural killer cell, a granulocyte, a neutrophil, an eosinophil, a basophil, a mast cell, a monocyte, a dendritic cell, a plasmacytoid dendritic cell, an NKT cell, and a macrophage;

(d) the antibody increases the percentage of polyfunctional (IFN γ + TNF α +) T cells;

(e) the antibody, when bound to activated regulatory T cells, binds to activating Fc gamma receptors selected from the group consisting of CD16, CD32A and CD64 to a greater extent than the antibody, when bound to activated effector T cells, binds to the activating Fc gamma receptors selected from the group consisting of CD16, CD32A and CD64;

optionally wherein the activating Fc gamma receptor is expressed on a cell selected from the group consisting of myeloid-derived effector cells and lymphocyte-derived effector cells;

(f) the antibody increases surface expression of OX40, PD-1, or a combination thereof in activated T cells;

(g) the antibody binds to human GITR with a K_D in the range of about 0.5 nM to 5 nM;
and/or

(h) the antibody further comprises a detectable label.

56. The antibody of any one of claims 1-55, wherein:

(a) the antibody partially inhibits human GITR ligand (GITRL) from binding to human GITR; optionally wherein the antibody inhibits 50% to 70% of human GITRL binding to human GITR;

(b) the antibody at a concentration of 1000ng/ml inhibits less than 80% of 0.5nM of human GITRL from binding to human GITR coupled to beads at a concentration of 5pg/ml/bead relative to the binding of 0.5nM human GITRL to the GITR coupled beads at a concentration of 5pg/ml/bead in the absence of the anti-GITR antibody in a suspension array assay; optionally wherein the antibody inhibits 50% to 70% of human GITRL binding to human GITR; or

(c) at least 20% of the amount of human GITRL that binds to human GITR in the absence of the antibody binds to human GITR in the presence of the antibody in an assay comprising the following steps:

- (a) coupling human GITR to beads at a concentration of 5pg/ml/bead;
- (b) incubating the human GITR coupled beads at a concentration of 40 beads/ μ l with or without 1000ng/ml of the antibody in a well;
- (c) adding labeled human GITRL to the well to obtain a final concentration of 0.5nM of the human GITRL and 20 beads/ μ l of the GITR coupled beads; and
- (d) detecting the labeled human GITRL bound to the human GITR coupled beads;

optionally wherein the antibody inhibits 50% to 70% of human GITRL binding to human GITR;

optionally wherein 30% to 50% of the amount of human GITRL that binds to human GITR in the absence of the antibody binds to human GITR in the presence of the antibody.

57. An isolated antibody that specifically binds to human GITR and partially inhibits human GITR ligand (GITRL) from binding to human GITR;

optionally wherein the antibody at a concentration of 1000ng/ml inhibits less than 80% of 0.5nM of human GITRL from binding to human GITR coupled to beads at a concentration of 5pg/ml/bead relative to the binding of 0.5nM GITRL to the GITR coupled beads at a concentration of 5pg/ml/bead in the absence of the anti-GITR antibody in a suspension array assay, optionally wherein the antibody inhibits 50% to 70% of human GITRL binding to human GITR; or

optionally wherein at least 20% of the amount of human GITRL that binds to human GITR in the absence of the antibody binds to human GITR in the presence of the antibody in an assay comprising the following steps:

- (a) coupling human GITR to beads at a concentration of 5pg/ml/bead;
- (b) incubating the human GITR coupled beads at a concentration of 40 beads/ μ l with or without 1000ng/ml of the antibody in a well;
- (c) adding labeled human GITRL to the well to obtain a final concentration of 0.5nM of the human GITRL and 20 beads/ μ l of the GITR coupled beads; and
- (d) detecting the labeled human GITRL bound to the human GITR coupled beads;

optionally wherein 30% to 50% of the amount of human GITRL that binds to human GITR in the absence of the antibody binds to human GITR in the presence of the antibody.

58. An isolated antibody that specifically binds to human GITR, wherein the antibody is capable of inducing, activating, or enhancing an activity of GITR or NF- κ B in a cell in the absence of TCR triggering.

59. An isolated antibody that binds to the same epitope of human GITR as the antibody of any one of claims 1-58.

60. The isolated antibody of any one of claims 1-59, wherein:

(a) the antibody binds to human GITR comprising residues 26-241 of SEQ ID NO: 701, wherein the binding between the antibody and a variant GITR is substantially weakened relative to the binding between the antibody and the human GITR, and wherein the variant GITR

comprises residues 26-241 of SEQ ID NO: 701 except for an amino acid substitution selected from the group consisting of D60A and G63A;

(b) the antibody binds to an epitope comprising residues 60-63 of SEQ ID NO: 701;

(c) the antibody binds to at least one residue within the amino acid sequence set forth by residues 60-63 of SEQ ID NO: 701;

(d) the antibody binds an epitope of the human GITR comprising at least one of residues 60 or 63 of SEQ ID NO: 701;

(e) the antibody specifically binds to each of i) human GITR, wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701 and ii) a variant of cynomolgus GITR, the variant comprising residues 26-234 of SEQ ID NO: 699, wherein the antibody does not specifically bind to cynomolgus GITR comprising residues 26-234 of SEQ ID NO: 704; and/or

(f) the antibody exhibits, as compared to binding to human GITR, reduced or absent binding to a protein identical to human GITR except for the presence of a D60A or G63A amino acid substitution.

61. An isolated antibody that specifically binds to human GITR, wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701, wherein the binding between the antibody and a variant GITR is substantially weakened relative to the binding between the antibody and the human GITR, and wherein the variant GITR comprises residues 26-241 of SEQ ID NO: 701 except for an amino acid substitution selected from the group consisting of D60A and G63A.

62. An isolated antibody that specifically binds to human GITR, wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701, and wherein the antibody binds to an epitope comprising residues 60-63 of SEQ ID NO: 701.

63. An isolated antibody that specifically binds to human GITR, wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701, and wherein the antibody binds to at least one residue within the amino acid sequence set forth by residues 60-63 of SEQ ID NO: 701.

64. An isolated antibody that specifically binds to human GITR,

wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701, and wherein the antibody binds an epitope of the human GITR comprising at least one of residues 60 or 63 of SEQ ID NO: 701; or

wherein the antibody exhibits, as compared to binding to human GITR, reduced or absent binding to a protein identical to human GITR except for the presence of a D60A or G63A amino acid substitution.

65. An isolated antibody that specifically binds to each of i) human GITR, wherein the human GITR comprises residues 26-241 of SEQ ID NO: 701 and ii) a variant of cynomolgus GITR, the variant comprising residues 26-234 of SEQ ID NO: 699, wherein the antibody does not specifically bind to cynomolgus GITR comprising residues 26-234 of SEQ ID NO: 704.

66. An isolated antibody that competes with the antibody of any one of claims 1-65 for binding to human GITR to the extent that the antibody of any one of claims 1-65 self-competes for binding to human GITR.

67. An isolated antibody that competes with the antibody of any one of claims 1-66 for binding to human GITR, wherein:

(1) the antibody competes for binding in an assay comprising the following steps:

- (a) incubating GITR-transfected cells with the antibody in unlabeled form in a container;
- (b) adding the antibody of any one of claims 1-66 in labeled form to the cells in the container and incubating the cells in the container; and
- (c) detecting the binding of the antibody of any one of claims 1-66 in labeled form to the cells; or

(2) the competition is exhibited as reduced binding of the antibody to human GITR by more than 80%.

68. An isolated nucleic acid molecule encoding a VH and/or a VL, or a heavy chain and/or a light chain, of the antibody of any one of claims 1-67.

69. An isolated vector comprising the nucleic acid molecule of claim 68.

70. A host cell comprising the isolated nucleic acid molecule of claim 68 or the isolated vector of claim 69;

optionally wherein the host cell is selected from the group consisting of *E. coli*, *Pseudomonas*, *Bacillus*, *Streptomyces*, yeast, CHO, YB/20, NS0, PER-C6, HEK-293T, NIH-3T3, HeLa, BHK, Hep G2, SP2/0, R1.1, B-W, L-M, COS 1, COS 7, BSC1, BSC40, BMT10 cell, plant cell, insect cell, and human cell in tissue culture.

71. A method of producing an antibody that binds to human GITR, comprising culturing the host cell of claim 70 so that the nucleic acid molecule is expressed and the antibody is produced.

72. An isolated antibody that specifically binds to human GITR and is encoded by the isolated nucleic acid molecule of claim 68.

73. A pharmaceutical composition comprising the antibody of any one of claims 1-67 and 72, the nucleic acid molecule of claim 68, the vector of claim 69, or the host cell of claim 70, and a pharmaceutically acceptable carrier or excipient.

74. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for modulating an immune response in a subject; optionally wherein the medicament enhances or induces the immune response of the subject.

75. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for enhancing the expansion of T cells and T cell effector function in a subject.

76. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for enhancing the expansion of CD8⁺ T cells in a subject.

77. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for activating T cells independent of TCR triggering in a subject.

78. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for inducing, activating, or enhancing an activity of NF- κ B independent of TCR triggering in a subject.

79. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for increasing the percentage of polyfunctional (IFN γ + TNF α +) T cells in a subject.

80. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for increasing surface expression of OX40, PD-1, or a combination thereof, in activated T cells in a subject.

81. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for treating cancer in a subject;

optionally wherein the cancer is squamous cell cancer, small-cell lung cancer, non-small cell lung cancer, gastrointestinal cancer, Hodgkin's or non-Hodgkin's lymphoma, pancreatic cancer, glioblastoma, glioma, cervical cancer, ovarian cancer, liver cancer, bladder cancer, breast cancer, colon cancer, colorectal cancer, endometrial carcinoma, myeloma, salivary gland carcinoma, kidney cancer, basal cell carcinoma, melanoma, prostate cancer, vulval cancer, thyroid cancer, testicular cancer, esophageal cancer, a type of head or neck cancer, desmoplastic melanoma, inflammatory breast cancer, thymoma, rectal cancer, anal cancer, or surgically treatable or non-surgically treatable brain stem glioma.

82. The use of claim 81, wherein the subject is further to be administered an inhibitor of indoleamine-2,3-dioxygenase (IDO);

optionally wherein the inhibitor is selected from the group consisting of epacadostat, F001287, indoximod, and NLG919.

83. Use of an effective amount of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for treating a viral infection in a subject;

optionally wherein the viral infection is a viral infection caused by a human papilloma virus (HPV), a Herpes simplex or other herpes virus, hepatitis B virus (HBV), hepatitis C virus (HCV) or other hepatitis virus, measles virus, HIV, or Epstein Barr virus (EBV),

optionally wherein the viral infection is meningitis, encephalitis, dengue or smallpox.

84. Use of an effective amount of the antibody of any one of claims 1-67 and 72 in combination with another therapeutic agent in the manufacture of a medicament for treating cancer in a subject;

optionally wherein the other therapeutic agent is an inhibitor of IDO, wherein the antibody and the inhibitor of IDO are to be administered in different pharmaceutical compositions and as separate dosage forms; optionally wherein the inhibitor is selected from epacadostat, F001287, indoximod and NLG919;

optionally wherein the other therapeutic agent is a checkpoint targeting agent;

optionally wherein the checkpoint targeting agent is selected from the group consisting of an antagonist of PD-1, an antagonist of PD-L1, an antagonist of PD-L2, an antagonist of CTLA-4, an antagonist of TIM-3, an antagonist of LAG-3, and an agonist of OX40;

optionally wherein the antagonist of PD-1 or the agonist of OX40 is to be administered subsequent to, simultaneously with, or prior to the administration of the antibody; and

optionally wherein the checkpoint targeting agent is selected from the group consisting of an antagonist anti-PD-1 antibody, an antagonist anti-PD-L1 antibody, an antagonist anti-PD-L2 antibody, an antagonist anti-CTLA-4 antibody, an antagonist anti-TIM-3 antibody, an antagonist anti-LAG-3 antibody, and an agonist anti-OX40 antibody; or

optionally wherein the other therapeutic agent is an anti-CD25 antibody;
optionally wherein the anti-CD25 antibody is to be administered
subsequent to, simultaneously with, or prior to the administration of the antibody.

85. Use of an agent in the manufacture of a medicament for treating cancer in a subject who has received an anti-GITR antibody, wherein:

- (a) the agent is an anti-PD-1 antibody, wherein the anti-PD-1 antibody is to be administered at a time at which the anti-GITR antibody has increased expression of PD-1 in the subject relative to expression of PD-1 in the subject at the time of the administering;
- (b) the agent is an anti-OX40 antibody, wherein the anti-OX40 antibody is to be administered at a time at which the anti-GITR antibody has increased expression of OX40 in the subject relative to expression of OX40 in the subject at the time of the administering;

optionally wherein the anti-GITR antibody induces, activates or enhances an activity of GITR.

86. Use of an anti-GITR antibody and a second antibody in the manufacture of a medicament for treating cancer in a subject, wherein:

- (a) the second antibody is an anti-PD-1 antibody, wherein the anti-GITR antibody increases expression of PD-1 in the subject relative to expression of PD-1 in the subject at the time of administration of the anti-GITR antibody, and wherein the anti-PD-1 antibody is to be administered to the subject when expression of PD-1 is increased; or
- (b) the second antibody is an anti-OX40 antibody, wherein the anti-GITR antibody increases expression of OX40 in the subject relative to expression of OX40 in the subject at the time of administration of the anti-GITR antibody, and wherein the anti-OX40 antibody is to be administered to the subject when expression of OX40 is increased;

optionally wherein the anti-GITR antibody induces, activates or enhances an activity of GITR.

87. Use of an effective amount of an isolated antibody that specifically binds to human GITR in the manufacture of a medicament for treating cancer in a subject, wherein the antibody is capable

of inducing, activating, or enhancing an activity of GITR or NF- κ B in a cell in the absence of TCR triggering.

88. The use of any one of claims 74-87, wherein the subject is human.

89. Use of the antibody of any one of claims 1-67 and 72 in the manufacture of a medicament for treating cancer or a viral infection in a first subject, wherein the antibody is incubated with T cells *ex vivo*; and wherein the T cells are to be infused into the first subject;

optionally wherein the T cells were isolated from a second subject, wherein the T cells are autologous or allogeneic to the first subject;

optionally wherein, prior to incubation:

- (1) the T cells are assayed for cell surface expression of GITR; and
- (2) if cell surface expression of GITR is not detected above a threshold value, GITR expression is induced on the surface of the T cells by incubating the T cells with a T cell receptor (TCR) complex stimulating agent; or

optionally wherein, prior to, simultaneously with, or subsequent to, the incubation, the T cells are to be incubated with a TCR complex stimulating agent; and

optionally wherein the first subject is to be administered an anti-cancer agent; optionally wherein the anti-cancer agent is a vaccine; optionally wherein the vaccine comprises a heat shock protein peptide complex (HSPPC) comprising a heat shock protein complexed with an antigenic peptide; and optionally wherein the heat shock protein is gp96 protein and is complexed with a tumor-associated antigenic peptide, wherein the HSPPC is derived from a tumor obtained from the first subject.

90. Use of the antibody of any one of claims 1-67 and 72 in the manufacture of a medicament for enhancing the expansion of T cells and T cell effector function, wherein the antibody is incubated *ex vivo* with T cells;

optionally wherein the T cells were isolated from a first subject;

optionally wherein, prior to, simultaneously with, or subsequent to the incubation, the T cells are to be incubated with a TCR complex stimulating agent; and/or

optionally wherein the T cells which were incubated with the antibody are to be infused into a second subject, wherein the T cells are autologous or allogeneic to the second subject.

91. Use of the antibody of any one of claims 1-67 and 72 in the manufacture of a medicament for enhancing the stimulation of T cells, wherein the antibody is incubated with T cells *ex vivo* that have been stimulated with a TCR complex stimulating agent;

optionally wherein the T cells were isolated from a first subject; and/or

optionally wherein the T cells that were incubated with the antibody are to be infused into a second subject, wherein the T cells are autologous or allogeneic to the second subject.

92. The use of any one of claims 89-91, wherein the first subject and/or second subject is human.

93. A method for activating T cells, comprising incubating *ex vivo* T cells with a TCR complex stimulating agent and the antibody of any one of claims 1-67 and 72;

optionally wherein the T cells were isolated from a first subject; and/or

optionally wherein the T cells that were incubated with the antibody are to be infused into a second subject, wherein the T cells are autologous or allogeneic to the second subject.

94. The method of claim 93, wherein the first subject and/or second subject is human.

95. Use of the antibody of any one of claims 1-67 and 72 or the pharmaceutical composition of claim 73 in the manufacture of a medicament for preferential expansion of effector T-cells over the expansion of T-regulatory cells in a subject; optionally wherein the subject is human.

96. A method of identifying an anti-GITR antibody that is capable of inducing, activating, or enhancing an activity of GITR in the absence of a TCR agonist, comprising contacting a cell expressing GITR with an anti-GITR antibody in the absence of a TCR agonist and measuring GITR activity, wherein increased GITR activity compared to GITR activity in the absence of the

anti-GITR antibody indicates the anti-GITR antibody is capable of inducing, activating, or enhancing an activity of GITR in the absence of a TCR agonist;

optionally wherein the GITR activity is assessed by:

- (a) measuring NF- κ B activity;
- (b) measuring activation of canonical and non-canonical NF- κ B pathways;
- (c) measuring activation of TRAF adapter mediated signaling pathways,
wherein the TRAF adapter is selected from the group consisting of
TRAF1, TRAF2, TRAF3, TRAF4, and TRAF5; or
- (d) measuring activation of MAPK/ERK pathway; and

optionally wherein the anti-GITR antibody increases the GITR activity at least two-fold compared to the GITR activity in the absence of the anti-GITR antibody; and/or

optionally wherein the cell is a T cell, or wherein the cell is not a T cell.