CLAIMS

1. Use of a radiolabeled antibody conjugate comprising an antibody or antigen-binding fragment thereof that binds lymphocyte activation gene-3 (LAG3) and a positron emitter, wherein the antibody or antigen-binding fragment thereof comprises three heavy chain complementarity determining regions (HCDRs) (HCDR1, HCDR2 and HCDR3) within a heavy chain variable region (HCVR) amino acid sequence of SEQ ID NO: 418 and three light chain complementarity determining regions (LCDRs) (LCDR1, LCDR2 and LCDR3) within a light chain variable region (LCVR) amino acid sequence of SEQ ID NO: 426, in the manufacture of a medicament for imaging a tissue that expresses LAG3, wherein the radiolabeled antibody conjugate is to be administered to the tissue; and LAG3 expression is to be visualized by positron emission tomography (PET) imaging.

2. The use of claim 1, wherein the radiolabeled antibody conjugate further comprises a chelating moiety.

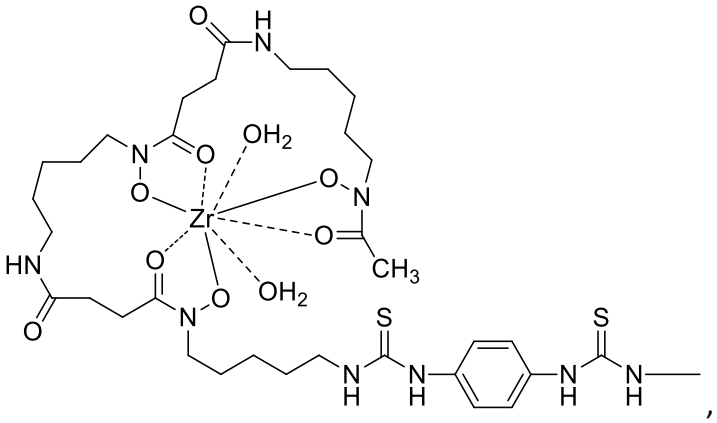
3. The use of claim 2, wherein the antibody or antigen-binding fragment thereof is covalently bonded to one or more moieties of Formula (**A**):

$$\text{-L-M}_z$$

(**A**)

wherein L is the chelating moiety; M is the positron emitter; and z, independently at each occurrence, is 0 or 1; and wherein at least one of z is 1.

4. The use of claim 3, wherein -L-M is



and wherein Zr is the positron emitter ^{89}Zr.

5. The use of claim 3 or claim 4, wherein the antibody or antigen-binding fragment thereof is covalently bonded to one, two, or three moieties of Formula (**A**).

6. The use of any one of claims 2 through 5, wherein the chelating moiety comprises desferrioxamine.

7. The use of any one of claims 1 through 6, wherein the positron emitter is ^{89}Zr.

8. The use of any one of claims 1 through 7, wherein the antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

9. The use of any one of claims 1 through 8, wherein the antibody or antigen-binding fragment thereof comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.

10. Use of a radiolabeled antibody conjugate comprising an antibody or antigen-binding fragment thereof that binds LAG3 and a positron emitter, wherein the antibody or antigen-binding fragment thereof comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418 and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426, in the

manufacture of a diagnostic reagent for determining that a cancer comprises LAG3-positive cells, wherein localization of the radiolabeled antibody conjugate in the cancer in a subject is to be imaged by positron emission tomography (PET) imaging, and wherein presence of the radiolabeled antibody conjugate in the cancer indicates that the cancer comprises LAG3-positive cells.

11. The use of claim 10, wherein the radiolabeled antibody conjugate further comprises a chelating moiety.

12. The use of claim 11, wherein the chelating moiety comprises desferrioxamine.

13. The use of any one of claims 10 through 12, wherein the positron emitter is ^{89}Zr.

14. The use of any one of claims 10 through 13, wherein the antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

15. The use of any one of claims 10 through 14, wherein the antibody or antigen-binding fragment thereof comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.

16. The use of any one of claims 10 through 15, wherein upon determining that the cancer comprises LAG3-positive cells, the cancer is further subject to an anti-cancer therapy selected from the group consisting of an inhibitor of LAG3, an inhibitor of the PD-1/PD-L1 signaling axis, a CTLA-4 inhibitor, a TIM3 inhibitor, a BTLA inhibitor, a TIGIT inhibitor, a CD47 inhibitor, a GITR inhibitor, an antagonist of another T cell co-inhibitor or ligand, an indoleamine-2,3-dioxygenase (IDO) inhibitor, a vascular endothelial growth factor (VEGF) antagonist, an Ang2 inhibitor, a transforming growth factor beta (TGFβ) inhibitor, an epidermal growth factor receptor (EGFR) inhibitor, a CD20 inhibitor, an antibody to a tumor-specific antigen, a cancer vaccine, a bispecific antibody, a cytotoxin, a chemotherapeutic agent, cyclophosphamide, radiotherapy,

an IL-6R inhibitor, an IL-4R inhibitor, an IL-10 inhibitor, IL-2, IL-7, IL-21, IL-15, and an antibody-drug conjugate (ADC).

17. The use of any one of claims 10 through 15, wherein upon determining that the cancer comprises LAG3-positive cells, the cancer is further subject to an anti-cancer therapy selected from the group consisting of an anti-LAG3 antibody, REGN2810, BGB-A317, nivolumab, pidilizumab, pembrolizumab, atezolizumab, avelumab, durvalumab, MDX-1105, REGN3504, ipilimumab, an anti-CD-28 antibody, an anti-2B4 antibody, an anti-LY108 antibody, an anti-LAIR1 antibody, an anti-ICOS antibody, an anti-CD160 antibody, an anti-VISTA antibody, aflibercept, bevacizumab, ranibizumab, sunitinib, sorafenib, pazopanib, nesvacumab, erlotinib, cetuximab, rituximab, an anti-CA9 antibody, an anti-CA125 antibody, an anti-melanoma-associated antigen 3 (MAGE3) antibody, an anti-carcinoembryonic antigen (CEA) antibody, an anti-vimentin antibody, an anti-tumor-M2-PK antibody, an anti-prostate-specific antigen (PSA) antibody, an anti-mucin-1 antibody, an anti-MART-1 antibody, an anti-CA19-9 antibody, Bacillus Calmette-Guerin, a CD3×CD20 bispecific antibody, a PSMA×CD3 bispecific antibody, dacarbazine, temozolomide, cyclophosphamide, docetaxel, doxorubicin, daunorubicin, cisplatin, carboplatin, gemcitabine, methotrexate, mitoxantrone, oxaliplatin, paclitaxel, vincristine, radiotherapy, sarilumab, dupilumab, anti-CD19-DM4 ADC, and anti-DS6-DM4 ADC.

18. The use of any one of claims 10 through 15, wherein upon determining that the cancer comprises LAG3-positive cells, the cancer is further subject to an anti-cancer therapy selected from the group consisting of an anti-LAG3 antibody or antigen-binding fragment thereof, an anti-PD-1 antibody or antigen-binding fragment thereof, and an anti-PD-L1 antibody or antigen-binding fragment thereof.

19. The use of claim 18, wherein the anti-cancer therapy is an anti-LAG3 antibody or antigen-binding fragment thereof comprising three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418, and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426.

20. The use of claim 18 or claim 19, wherein the anti-cancer therapy is an anti-LAG3 antibody or antigen-binding fragment thereof, and wherein the anti-LAG3 antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the

HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

21. The use of any one of claims 18 through 20, wherein the anti-cancer therapy is an anti-LAG3 antibody or antigen-binding fragment thereof, and wherein the anti-LAG3 antibody or antigen-binding fragment thereof comprises the HCVR of SEQ ID NO: 418 and the LCVR of SEQ ID NO: 426.

22. The use of claim 18, wherein the anti-cancer therapy is an anti-PD-1 antibody or antigen-binding fragment thereof selected from the group consisting of REGN2810, nivolumab, and pembrolizumab.

23. The use of claim 18, wherein the anti-cancer therapy is an anti-PD-L1 antibody or antigen-binding fragment thereof selected from the group consisting of atezolizumab, avelumab, and durvalumab.

24. The use of claim 18, wherein the anti-cancer therapy is an anti-LAG3 antibody or antigen-binding fragment thereof and the anti-LAG3 antibody or antigen-binding fragment thereof is to be administered in combination with a second anti-cancer therapy.

25. The use of claim 24, wherein the second anti-cancer therapy is selected from the group consisting of an inhibitor of the PD-1/PD-L1 signaling axis, a CTLA-4 inhibitor, a TIM3 inhibitor, a BTLA inhibitor, a TIGIT inhibitor, a CD47 inhibitor, a GITR inhibitor, an antagonist of another T cell co-inhibitor or ligand, an indoleamine-2,3-dioxygenase (IDO) inhibitor, a vascular endothelial growth factor (VEGF) antagonist, an Ang2 inhibitor, a transforming growth factor beta (TGFβ) inhibitor, an epidermal growth factor receptor (EGFR) inhibitor, a CD20 inhibitor, an antibody to a tumor-specific antigen, a cancer vaccine, a bispecific antibody, a cytotoxin, a chemotherapeutic agent, cyclophosphamide, radiotherapy, an IL-6R inhibitor, an IL-4R inhibitor, an IL-10 inhibitor, IL-2, IL-7, IL-21, IL-15, and an antibody-drug conjugate (ADC).

26. The use of claim 24, wherein the second anti-cancer therapy is selected from the group consisting of a further anti-LAG3 antibody, REGN2810, BGB-A317, nivolumab, pidilizumab,

pembrolizumab, atezolizumab, avelumab, durvalumab, MDX-1105, REGN3504, ipilimumab, an anti-CD-28 antibody, an anti-2B4 antibody, an anti-LY108 antibody, an anti-LAIR1 antibody, an anti-ICOS antibody, an anti-CD160 antibody, an anti-VISTA antibody, aflibercept, bevacizumab, ranibizumab, sunitinib, sorafenib, pazopanib, nesvacumab, erlotinib, cetuximab, rituximab, an anti-CA9 antibody, an anti-CA125 antibody, an anti-melanoma-associated antigen 3 (MAGE3) antibody, an anti-carcinoembryonic antigen (CEA) antibody, an anti-vimentin antibody, an anti-tumor-M2-PK antibody, an anti-prostate-specific antigen (PSA) antibody, an anti-mucin-1 antibody, an anti-MART-1 antibody, an anti-CA19-9 antibody, Bacillus Calmette-Guerin, a CD3×CD20 bispecific antibody, a PSMA×CD3 bispecific antibody, dacarbazine, temozolomide, cyclophosphamide, docetaxel, doxorubicin, daunorubicin, cisplatin, carboplatin, gemcitabine, methotrexate, mitoxantrone, oxaliplatin, paclitaxel, vincristine, radiotherapy, sarilumab, dupilumab, anti-CD19-DM4 ADC, and anti-DS6-DM4 ADC.

27. The use of any one of claims 10 through 26, wherein the cancer is selected from the group consisting of blood cancer, brain cancer, renal cell cancer, ovarian cancer, bladder cancer, prostate cancer, breast cancer, hepatic cell carcinoma, bone cancer, colon cancer, non-small-cell lung cancer, squamous cell carcinoma of head and neck, colorectal cancer, mesothelioma, B cell lymphoma, and melanoma.

28. Use of a radiolabeled antibody conjugate comprising an antibody or antigen-binding fragment thereof that binds LAG3 and a positron emitter, wherein the antibody or antigen-binding fragment thereof comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418 and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426, in the manufacture of a medicament for detecting LAG3-expressing lymphocytes in a subject.

29. The use of claim 28, wherein the subject suffers from cancer, a viral infection, a bacterial infection, a fungal infection, or a parasitic infection.

30. The use of claim 28 or claim 29, wherein the radiolabeled antibody conjugate further comprises a chelating moiety.

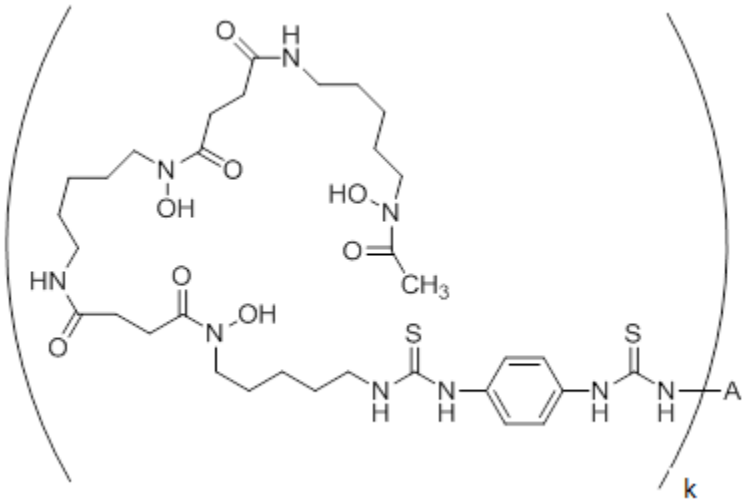
31. The use of claim 30, wherein the chelating moiety comprises desferrioxamine.

32. The use of any one of claims 28 through 31, wherein the positron emitter is ^{89}Zr.

33. The use of any one of claims 28 through 32, wherein the antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

34. The use of any one of claims 28 through 33, wherein the antibody or antigen-binding fragment thereof comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.

35. A compound of Formula (III):



,

wherein A is an antibody or antigen-binding fragment thereof that binds LAG3 and k is from 1-30, and

wherein the antibody or antigen-binding fragment thereof comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418, and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426.

36. The compound of claim 35, wherein the antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

37. The compound of claim 35 or claim 36, wherein the antibody or antigen-binding fragment thereof comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.

38. The compound of any one of claims 35 through 37, wherein k is greater than or equal to 1 but less than or equal to 2.

39. The compound of any one of claims 35 through 38, wherein k is 1 or 2.

40. Use of a radiolabeled antibody conjugate comprising an antibody or antigen-binding fragment thereof that binds LAG3 and a positron emitter, wherein the antibody or antigen-binding fragment thereof comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418 and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426, in the manufacture of a medicament for identifying T-cell exhaustion in a subject.

41. The use of claim 40, wherein the subject suffers from cancer, a viral infection, a bacterial infection, a fungal infection, or a parasitic infection.

42. The use of claim 40 or claim 41, wherein the radiolabeled antibody conjugate further comprises a chelating moiety.

43. The use of claim 42, wherein the chelating moiety comprises desferrioxamine.

44. The use of any one of claims 40 through 43, wherein the positron emitter is ^{89}Zr.

45. The use of any one of claims 40 through 44, wherein the antibody or antigen-binding fragment thereof comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424,

the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

46. The use of any one of claims 40 through 45, wherein the antibody or antigen-binding fragment thereof comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.

47. A composition comprising:

(i) a radiolabeled antibody conjugate comprising an antibody or antigen-binding fragment thereof that binds LAG3, desferrioxamine, and a ^{89}Zr positron emitter, wherein the antibody or antigen-binding fragment thereof comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418 and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426; and

(ii) LAG3 expressing T cells.

48. A method for making a radiolabeled anti-LAG3 binding protein, comprising:

loading a positron emitter onto a conjugate comprising an anti-LAG3 binding protein modified with a chelating agent, thereby forming the radiolabeled anti-LAG3 binding protein,

wherein the anti-LAG3 binding protein comprises three HCDRs within an HCVR amino acid sequence of SEQ ID NO: 418 and three LCDRs within an LCVR amino acid sequence of SEQ ID NO: 426.

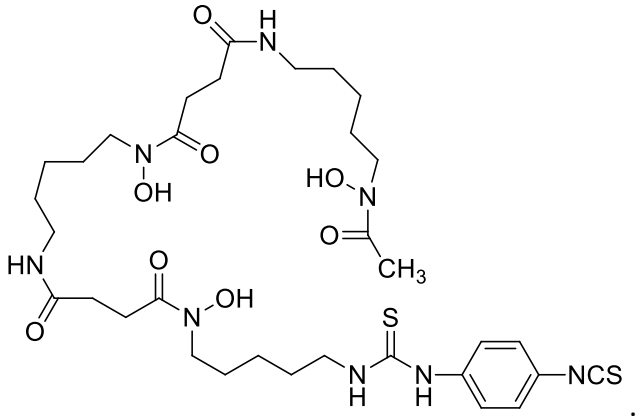
49. The method of claim 48, wherein the method further comprises:

forming the conjugate by reacting the anti-LAG3 binding protein with the chelating agent, wherein the chelating agent comprises a chelating moiety and a reactive moiety for conjugation to the anti-LAG3 binding protein, and

wherein the step of forming the conjugate occurs prior to loading the positron emitter.

50. The method of claim 49, wherein the reactive moiety is selected from the group consisting of an isothiocyantatobenzyl group, an n-hydroxysuccinimide ester, a 2,3,5,6 tetrafluorophenol ester, and an n-succinimidyl-*S*-acetylthioacetate moiety.

51. The method of any one of claims 48 through 50, wherein the chelating agent has the following formula:



52. The method of any one of claims 48 through 50, wherein the chelating moiety of the chelating agent comprises desferrioxamine.

53. The method of any one of claims 48 through 50, wherein the chelating agent is p-isothiocyanatobenzyl-desferrioxamine.

54. The method of any one of claims 48 through 53, wherein the positron emitter is ^{89}Zr.

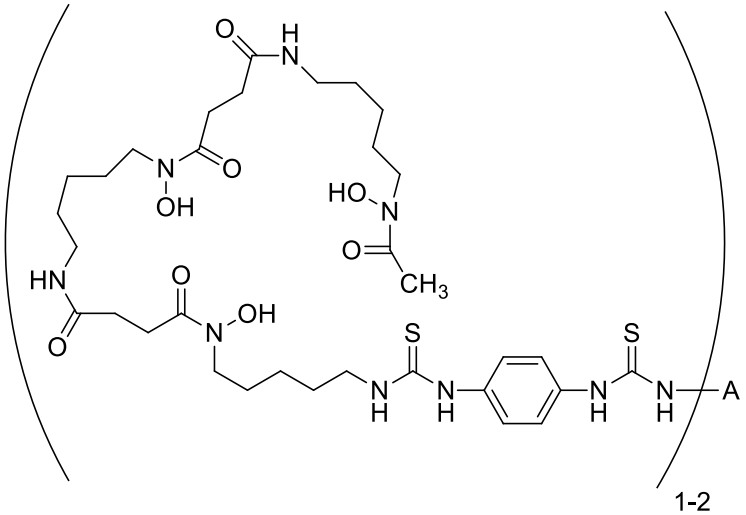
55. The method of any one of claims 48 through 53, wherein the positron emitter is an oxalic acid or other salt of ^{89}Zr.

56. The method of any one of claims 48 through 55, wherein the chelating agent-to-anti-LAG3-binding protein ratio is from one to four.

57. The method of any one of claims 48 through 55, wherein the chelating agent-to-anti-LAG3-binding protein ratio is from one to two.

58. The method of any one of claims 48 through 55, wherein the chelating agent-to-anti-LAG3-binding protein ratio is 1.48.

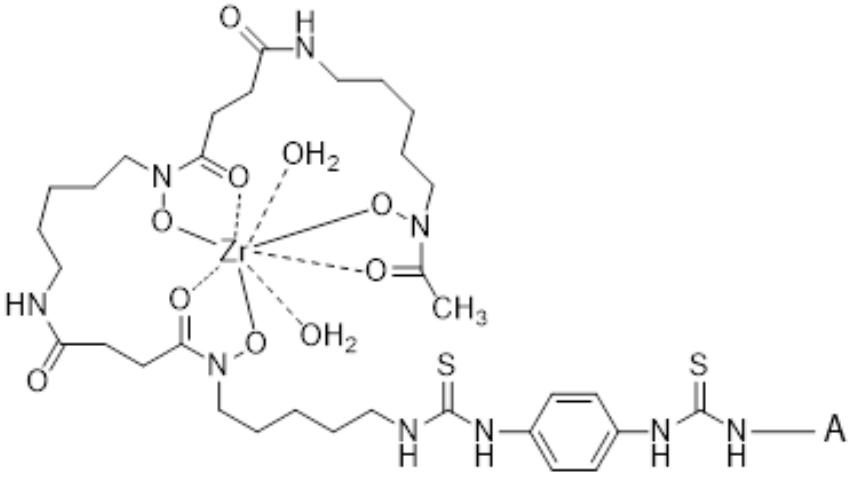
59. The method of any one of claims 48 through 58, wherein the conjugate has a formula:



1-2 (III),

or a salt thereof, wherein -A is the anti-LAG3 binding protein.

60. The method of any one of claims 48 through 59, wherein the radiolabeled anti-LAG3 binding protein is:



,

wherein A is the anti-LAG3 binding protein and Zr is the positron emitter ^{89}Zr.

61. The method of any one of claims 48 through 60, wherein the step of loading the positron emitter comprises incubating the conjugate with the positron emitter.

62. The method of any one of claims 48 through 61, wherein the anti-LAG3 binding protein has one or more properties selected from the group consisting of:

(a) binds monomeric human LAG3 with a binding dissociation equilibrium constant (K_D) of less than 8 nM as measured in a surface plasmon resonance assay at 37°C;

(b) binds monomeric human LAG3 with a K_D less than 10 nM in a surface plasmon resonance assay at 25°C;

(c) binds dimeric human LAG3 with a K_D of less than 1 nM as measured in a surface plasmon resonance assay at 37°C; and

(d) binds dimeric human LAG3 with a K_D less than 1.1 nM in a surface plasmon resonance assay at 25°C.

63. The method of any one of claims 48 through 62, wherein the anti-LAG3 binding protein comprises the HCDR1 amino acid sequence of SEQ ID NO: 420, the HCDR2 amino acid sequence of SEQ ID NO: 422, the HCDR3 amino acid sequence of SEQ ID NO: 424, the LCDR1 amino acid sequence of SEQ ID NO: 428, the LCDR2 amino acid sequence of SEQ ID NO: 430, and the LCDR3 amino acid sequence of SEQ ID NO: 432.

64. The method of any one of claims 48 through 63, wherein the anti-LAG3 binding protein comprises the HCVR amino acid sequence of SEQ ID NO: 418 and the LCVR amino acid sequence of SEQ ID NO: 426.