

Claims

1. An isolated human heavy chain variable domain (V_H) single domain antibody that binds to human LAG-3 comprising a CDR3 sequence selected from SEQ ID No. 3 or 7 or a sequence with at least 70%, 80% or 90% homology thereto.
2. The isolated V_H single domain antibody according to claim 1 comprising a CDR1 as shown in SEQ ID No. 1 or a sequence with at least 70%, 80% or 90% homology thereto, a CDR2 as shown in SEQ ID No. 2 or a sequence with at least 70%, 80% or 90% homology thereto and a CDR3 as shown in SEQ ID No. 3 or a sequence with at least 70%, 80% or 90% homology thereto.
3. The isolated V_H single domain antibody according to claim 2 comprising a CDR1 as shown in SEQ ID No. 5 or a sequence with at least 70%, 80% or 90% homology thereto, a CDR2 as shown in SEQ ID No. 6 or a sequence with at least 70%, 80% or 90% homology thereto and a CDR3 as shown in SEQ ID No. 7 or a sequence with at least 70%, 80% or 90% homology thereto.
4. The single domain antibody according to a preceding claim comprising a sequence selected from SEQ ID Nos 4 or 8 or a sequence with at least 70%, 80% or 90% homology thereto.
5. The isolated single domain antibody according to a preceding claim wherein said single domain antibody is conjugated to a toxin, enzyme, radioisotope, half-life extending moiety, label, therapeutic molecule or other chemical moiety.
6. The isolated single domain antibody according to claim 6 wherein said half-life extending moiety is selected from the group consisting of an albumin binding moiety, a transferrin binding moiety, a polyethylene glycol molecule, a recombinant polyethylene glycol molecule, human serum albumin, a fragment of human serum albumin, and an albumin binding peptide or single domain antibody that binds to human serum albumin.
7. The isolated single domain antibody according to a preceding claim obtained or obtainable from a transgenic rodent that expresses a transgene comprising human V, D and J regions.
8. The isolated single domain antibody according to claim 7 wherein said rodent does not produce any functional endogenous light and heavy chains.
9. An isolated binding agent comprising a first (V_H) single domain antibody that binds to human LAG-3 and a second (V_H) single domain antibody that binds to human LAG-3.
10. An isolated binding agent according to claim 9 wherein the first and/or second (V_H) single domain antibody that binds to human LAG-3 is selected from an isolated single domain antibody according to any of claims 1 to 8.

11. An isolated binding agent according to claim 9 or a binding agent comprising the single domain antibody of any of claims 1 to 8 linked to a second functional moiety having a different binding specificity than said single domain antibody.
12. The isolated binding agent according to claim 11 wherein said second functional moiety does not bind to LAG-3.
13. The isolated binding agent according to any of claims 11 or 12 wherein said second binding molecule is an antibody or a fragment thereof.
14. The isolated binding agent according to claim 13 wherein said fragment is a Fab, Fab', F(ab')₂, dAb, Fd, Fv, or a single chain Fv fragment, a human heavy chain variable domain (V_H), or an isolated CDR.
15. The isolated binding agent according to claim 14 wherein said second functional moiety is a V_H single domain antibodies having SEQ ID No. 4 or SEQ ID No. 8.
16. A multispecific binding agent comprising a V_H single domain antibody according to any of claims 1 to 8 or a binding agent according to any of claims 9 to 15 and a second functional moiety that binds to PD-1.
17. The multispecific binding agent according to claim 16 wherein the second functional moiety that binds to PD-1 is an antibody or a fragment thereof.
18. The multispecific binding agent according to claim 17 wherein the fragment is a Fab, Fab', F(ab')₂, dAb, Fd, Fv, or a single chain Fv fragment, a V_H human heavy chain variable domain or an isolated CDR.
19. The multispecific binding agent according to any claims 16 to 18 wherein the second functional moiety that binds to PD-1 is a V_H single domain antibody.
20. The multispecific binding agent according to claim 19 wherein the V_H single domain antibody that binds PD-1 is selected from a V_H single domain antibody listed in table 5.
21. The multispecific binding agent according to claim 20 wherein the V_H single domain antibody that binds PD-1 is selected from V_H 2.63, 2.77 or 2.92.
22. The multispecific binding agent according to claim 21 comprising two V_H single domain antibodies that bind LAG-3.
23. The multispecific binding agent according to claim 22 wherein two V_H single domain antibodies that bind LAG-3 have SEQ ID NO. 8 or a sequence with at least 70%, 80% or 90% homology thereto.
24. The binding agent according to any of claims 9 to 23 conjugated to a toxin, enzyme, radioisotope, half-life extending moiety, therapeutic molecule or other chemical moiety.
25. The binding agent according to claim 24 wherein said half-life extending moiety is selected from the group consisting of an albumin binding moiety, a transferrin binding moiety, a polyethylene glycol molecule, a recombinant polyethylene glycol molecule,

- human serum albumin, a fragment of human serum albumin, and an albumin binding peptide or single domain antibody that binds to human serum albumin.
26. The binding agent according to any of claims 9 to 25 wherein the moieties are linked with a (G4S)_n linker wherein n=1 to 20.
27. A use of a single domain antibody according to any of claims 1 to 8 in a multivalent binding agent.
28. The use of a single domain antibody according to claim 27 wherein the multivalent binding agent is multispecific.
29. The use of a single domain antibody according to claim 28 wherein the multispecific binding agent binds to LAG-3 and PD-1.
30. An immunoconjugate comprising a single domain antibody according to any one of claims 1 to 8 or a binding agent according to any one of claims 9 to 26 linked to a therapeutic agent.
31. A pharmaceutical composition comprising a single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 and a pharmaceutical carrier.
32. A method for treating a cancer, an immune disease, neurological disease, inflammatory disease, allergy, transplant rejection, viral infection, immune deficiency or other immune system-related disease comprising administering a therapeutically effective amount of a comprising a single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 31.
33. Use of a single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 31 in the manufacture of a medicament for the treatment of a cancer, an immune disease, neurological disease, inflammatory disease, allergy, transplant rejection, infection, immune deficiency or other immune system-related disease.
34. A single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 31 for use as medicament.
35. A single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 34 for use in the treatment of a cancer, an immune disease, neurological disease, inflammatory disease, allergy, transplant rejection, viral infection, immune deficiency, and other immune system-related disease.

36. A method according to claim 32, a use according to claim 33, or a single domain antibody, a binding agent or pharmaceutical composition according to claim 31 wherein said cancer is selected from bone cancer, pancreatic cancer, skin cancer, cancer of the head or neck, cutaneous or intraocular malignant melanoma, uterine cancer, ovarian cancer, rectal cancer, cancer of the anal region, stomach cancer, testicular cancer, breast cancer, brain cancer, carcinoma of the fallopian tubes, carcinoma of the endometrium, carcinoma of the cervix, carcinoma of the vagina, carcinoma of the vulva, cancer of the esophagus, cancer of the small intestine, cancer of the endocrine system, cancer of the thyroid gland, cancer of the parathyroid gland, cancer of the adrenal gland, kidney cancer, sarcoma of soft tissue, cancer of the urethra, cancer of the bladder, renal cancer, lung cancer, non-small cell lung cancer, thymoma, urothelial carcinoma leukemia, prostate cancer, mesothelioma, adrenocortical carcinoma, lymphomas, such as such as Hodgkin's disease, non-Hodgkin's, gastric cancer, and multiple myelomas and other leukemias.
37. A method of modulating an immune response comprising administering a single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 23 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 31.
38. An isolated nucleic acid molecule comprising a nucleotide sequence encoding a single domain antibody according to any of claims 1 to 8 or a binding agent according to any of claims 9 to 26.
39. An isolated nucleic acid molecule according to claim 38 selected from SEQ ID Nos. 10 or 11.
40. A vector comprising a nucleic acid according to claim 38 or 39.
41. A host cell comprising a nucleic acid according to claim 35 or 36 or a vector according to claim 40.
42. A host cell according to claim 41 wherein said host cell is a bacterial, yeast, viral, plant or mammalian cell.
43. A method for producing a single domain antibody according to any one of claims 1 to 8 comprising expressing a nucleic acid encoding said binding molecule in a host cell and isolating the binding molecule from the host cell.
44. A kit comprising a single domain antibody according to any one of claims 1 to 8, a binding agent according to any of claims 9 to 26 or an immunoconjugate according to claim 30 or a pharmaceutical composition according to claim 31.
45. A method for detecting the presence of human LAG-3 in a test sample comprising contacting said sample with a single domain antibody according to any of claims 1 to 8

- and at least one detectable label and detecting binding of said single domain antibody to human LAG-3.
46. A combination therapy comprising administration of a single domain antibody according to any one of claims 1 to 8 together with a second therapeutic agent.
47. A method of simultaneously inhibiting immune checkpoints PD-1 and LAG-3, comprising administering a binding agent of any of claims 16 to 26.
48. A method of modifying or blocking the interaction of LAG-3 and galectin-3, Lselectin or alpha synuclein comprising administering a single domain antibody of any of claims 1 to 8 or a binding agent of any of claims 9 to 26.
49. A method of modifying or blocking the interaction of LAG-3 and galectin-3, Lselectin or alpha synuclein comprising contacting LAG-3 with a single domain antibody of any of claims 1 to 8 or a binding agent of any of claims 9 to 26.
50. A CAR comprising a single domain antibody according to any one of claims 1 to 8 or a binding agent according to any one of claims 9 to 26.
51. A method for selectively targeting cells that display PD-1 and LAG-3 on their surface or increasing targeting of such cells compared to cells that display PD-1 on their surface comprising providing a binding agent according to any one of claims 9 to 26.
52. A method according to claim 51 wherein the method is in vitro, in vivo or ex vivo.
53. A method for identifying a subject that responds to therapy with a bispecific agent that binds to LAG-3 and PD-1 by measuring the protein or RNA expression of Cbl-b and/or c-Cbl in said subject or a sample from said subject exposed to the bispecific therapy.
54. The method of claim 53 wherein the therapy is with a binding agent according to any one of claims 9 to 26.