

WHAT IS CLAIMED IS:

1. An antibody heavy chain variable domain comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO:7.
2. The antibody heavy chain variable domain according to claim 1, wherein the amino acid sequence comprises:
 - a complementarity-determining region 1 (CDR1) sequence represented by the amino acid sequence of SEQ ID NO:48;
 - a complementarity-determining region 2 (CDR2) sequence represented by the amino acid sequence of SEQ ID NO:30; and
 - 10 a complementarity-determining region 3 (CDR3) sequence represented by the amino acid sequence of SEQ ID NO:44.
3. The antibody heavy chain variable domain according to claim 1, wherein the amino acid sequence comprises:
 - a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:29;
 - 15 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:30; and
 - a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:31.
4. The antibody heavy chain variable domain according to claim 1, wherein the amino acid sequence comprises:
 - a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:48;
 - 20 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:30; and
 - a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:71.
5. The antibody heavy chain variable domain according to claim 1, wherein the amino acid sequence comprises:
 - a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:29;
 - 25 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:30; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:77.

6. The antibody heavy chain variable domain according to claim 1, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:48;

5 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:30; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:78.

7. An antibody heavy chain variable domain comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO:1.

8. The antibody heavy chain variable domain according to claim 7, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:11;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:12; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:13.

9. The antibody heavy chain variable domain according to claim 7, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:45;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:12; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:68.

10. An antibody heavy chain variable domain comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO:3.

11. The antibody heavy chain variable domain according to claim 10, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:17;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:18; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:19.

12. The antibody heavy chain variable domain according to claim 10, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:46;

5 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:18; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:69.

13. An antibody heavy chain variable domain comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO:5.

14. The antibody heavy chain variable domain according to claim 13, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:23;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:24; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:25.

15. The antibody heavy chain variable domain according to claim 13, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:47;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:24; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:70.

16. An antibody heavy chain variable domain comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO:9.

17. The antibody heavy chain variable domain according to claim 16, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:35;

a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:36; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:37.

18. The antibody heavy chain variable domain according to claim 16, wherein the amino acid sequence comprises:

a CDR1 sequence represented by the amino acid sequence of SEQ ID NO:45;

5 a CDR2 sequence represented by the amino acid sequence of SEQ ID NO:36; and

a CDR3 sequence represented by the amino acid sequence of SEQ ID NO:72.

19. An antibody heavy chain comprising an antibody heavy chain variable domain according to any one of claims 1-18 and an amino acid sequence at least 90% identical to an antibody constant region.

10 20. The antibody heavy chain according to claim 19, wherein the antibody constant region is a human IgG constant region comprising hinge, CH2, and CH3 domains.

21. The antibody heavy chain according to claim 20, wherein the antibody constant region is a human IgG constant region further comprising a CH1 domain.

15 22. The antibody heavy chain according to any one of claims 19-21, wherein the antibody constant region is 90% identical to a IgG1 constant region.

23. The antibody heavy chain according to claim 22, wherein the amino acid sequence at least 90% identical to an antibody constant region differs from the amino acid sequence of an IgG1 constant region at Q347, Y349, L351, S354, E356, E357, K360, Q362, S364, T366, L368, K370, N390, K392, T394, D399, S400, D401, F405, Y407, K409, T411, or K439, or
20 any combination thereof.

24. The antibody heavy chain according to claim 23, wherein the amino acid sequence at least 90% identical to an antibody constant region differs from the amino acid sequence of an IgG1 constant region by a substitution selected from the group consisting of Q347E, Q347R, Y349S, Y349K, Y349T, Y349D, Y349E, Y349C, L351K, L351D, L351Y, S354C, E356K, E357Q, E357L, E357W, K360E, K360W, Q362E, S364K, S364E, S364H, S364D, T366V, T366I, T366L, T366M, T366K, T366W, T366S, L368E, L368A, L368D, K370S, N390D, N390E, K392L, K392M, K392V, K392F, K392D, K392E, T394F, D399R, D399K, D399V,
25

S400K, S400R, D401K, F405A, F405T, Y407A, Y407I, Y407V, K409F, K409W, K409D, T411D, T411E, K439D, and K439E, or any combination thereof.

25. An antigen-binding site comprising the antibody heavy chain variable domain according to any one of claims 1-6, and an antibody light chain variable domain comprising an amino acid sequence at least 90% identical to SEQ ID NO:8.
26. The antigen-binding site according to claim 25, wherein the light chain variable domain comprises a CDR1 sequence identical to the amino acid sequence of SEQ ID NO:32, a CDR2 sequence identical to the amino acid sequence of SEQ ID NO:33, and a CDR3 sequence identical to the amino acid sequence of SEQ ID NO:34.
27. The antigen-binding site according to claim 25 or 26 having a K_D of between 2 – 120 nM, as measured by surface plasmon resonance.
28. An antigen-binding site comprising the antibody heavy chain variable domain according to any one of claims 7-9, and an antibody light chain variable domain comprising an amino acid sequence at least 90% identical to SEQ ID NO:2.
29. The antigen-binding site according to claim 28, wherein the light chain variable domain comprises a CDR1 sequence identical to the amino acid sequence of SEQ ID NO:14, a CDR2 sequence identical to the amino acid sequence of SEQ ID NO:15, and a CDR3 sequence identical to the amino acid sequence of SEQ ID NO:16.
30. The antigen-binding site according to claim 28 or 29 having a K_D of between 5 – 500 nM, as measured by surface plasmon resonance.
31. An antigen-binding site comprising the antibody heavy chain variable domain according to any one of claims 10-12, and an antibody light chain variable domain comprising an amino acid sequence at least 90% identical to SEQ ID NO:4.
32. The antigen-binding site according to claim 31, wherein the light chain variable domain comprises a CDR1 sequence identical to the amino acid sequence of SEQ ID NO:20, a CDR2 sequence identical to the amino acid sequence of SEQ ID NO:21, and a CDR3 sequence identical to the amino acid sequence of SEQ ID NO:22.

33. The antigen-binding site according to claim 31 or 32 having a K_D of between 6 – 600 nM, as measured by surface plasmon resonance.
34. An antigen-binding site comprising the antibody heavy chain variable domain according to any one of claims 13-15, and an antibody light chain variable domain
5 comprising an amino acid sequence at least 90% identical to SEQ ID NO:6.
35. The antigen-binding site according to claim 34, wherein the light chain variable domain comprises a CDR1 sequence identical to the amino acid sequence of SEQ ID NO:26, a CDR2 sequence identical to the amino acid sequence of SEQ ID NO:27, and a CDR3 sequence identical to the amino acid sequence of SEQ ID NO:28.
- 10 36. The antigen-binding site according to claim 34 or 35 having a K_D of between 1 – 100 nM, as measured by surface plasmon resonance.
37. An antigen-binding site comprising the antibody heavy chain variable domain according to any one of claims 16-18, and an antibody light chain variable domain comprising an amino acid sequence at least 90% identical to SEQ ID NO:10.
- 15 38. The antigen-binding site according to claim 37, wherein the light chain variable domain comprises a CDR1 sequence identical to the amino acid sequence of SEQ ID NO:38, a CDR2 sequence identical to the amino acid sequence of SEQ ID NO:39, and a CDR3 sequence identical to the amino acid sequence of SEQ ID NO:40.
39. The antigen-binding site according to claim 37 or 38 having a K_D of between 6 – 600
20 nM, as measured by surface plasmon resonance.
40. A protein comprising:

an antigen-binding site according to any one of claims 25-39, wherein the antigen-binding site binds to NKG2D; and an additional antigen-binding site.
41. The protein according to claim 40, wherein the additional antigen-binding site binds
25 to a tumor-associated antigen.
42. The protein according to claim 41, wherein the tumor-associated antigen is selected from the group consisting of CD33, HER2, EpCAM, CD2, CD19, CD20, CD30, CD38, CD40, CD52, CD70, EGFR/ERBB1, IGF1R, HER3/ERBB3, HER4/ERBB4, MUC1, cMET,

SLAMF7, PSCA, MICA, MICB, TRAILR1, TRAILR2, MAGE-A3, B7.1, B7.2, CTLA4, and PD1.

43. The protein according to any one of claims 40-42, wherein the antigen-binding site binding NKG2D comprises a first antibody heavy chain variable domain, and the additional
5 antigen-binding site comprises a second antibody heavy chain variable domain; and

wherein the first antibody heavy chain variable domain is present on a first polypeptide further comprising a first antibody constant region, and the second antibody heavy chain variable domain is present on a second polypeptide further comprising a second antibody constant region.

- 10 44. The protein according to claim 43, wherein the first antibody constant region and the second antibody constant region form a complex capable of binding CD16.

45. The protein according to claim 43 or 44, wherein the first antibody constant region and the second antibody constant region each comprise hinge, CH2, and CH3 domains.

- 15 46. The protein according to any one of claims 43-45, wherein the first antibody constant region and the second antibody constant region each further comprise a CH1 domain.

47. The protein according to any one of claims 43-46, wherein the amino acid sequences of the first antibody constant region and the second antibody constant region are each at least 90% identical to human IgG1 constant region.

48. The protein according to claim 47, wherein:

- 20 the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at Q347, Y349, L351, S354, E356, E357, K360, Q362, S364, T366, L368, K370, K392, T394, D399, S400, D401, F405, Y407, K409, T411, or K439, or any combination thereof; and

- 25 the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at Q347, Y349, L351, S354, E356, E357, S364, T366, L368, K370, N390, K392, T394, D399, D401, F405, Y407, K409, T411, or K439, or any combination thereof.

49. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at position T366, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at T366, L368, or Y407, or
5 any combination thereof.

50. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at T366, L368, or Y407, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1
10 constant region at position T366.

51. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at E357, K360, Q362, S364, L368, K370, T394, D401, F405, or T411, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs
15 from the amino acid sequence of an IgG1 constant region at Y349, E357, S364, L368, K370, T394, D401, F405, or T411, or any combination thereof.

52. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at Y349, E357, S364, L368, K370, T394, D401, F405, or T411, or any combination thereof, and
20 wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at E357, K360, Q362, S364, L368, K370, T394, D401, F405, or T411, or any combination thereof.

53. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at L351, D399, S400, or Y407, or any combination thereof, and wherein the amino acid
25 sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at T366, N390, K392, K409, or T411, or any combination thereof.

54. The protein according to claim 47, wherein the amino acid sequence of the a first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
30 T366, N390, K392, K409, or T411, or any combination thereof, and wherein the amino acid

sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at L351, D399, S400, or Y407, or any combination thereof.

55. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
5 Q347, Y349, K360, or K409, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at Q347, E357, D399, or F405, or any combination thereof.

56. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
10 Q347, E357, D399, or F405, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at Y349, K360, Q347, or K409, or any combination thereof.

57. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
15 K370, K392, K409, or K439, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at D356, E357, or D399, or any combination thereof.

58. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
20 D356, E357, or D399, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at K370, K392, K409, or K439, or any combination thereof.

59. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
25 L351, E356, T366, or D399, or any combination thereof, and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at Y349, L351, L368, K392, or K409, or any combination thereof.

60. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region at
30 Y349, L351, L368, K392, or K409, or any combination thereof, and wherein the amino acid

sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region at L351, E356, T366, or D399, or any combination thereof.

61. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by an S354C substitution and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by a Y349C substitution.

62. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by a Y349C substitution and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by an S354C substitution.

63. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by K360E and K409W substitutions and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by O347R, D399V and F405T substitutions.

64. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by O347R, D399V and F405T substitutions and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by K360E and K409W substitutions.

65. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by a T366W substitutions and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by T366S, T368A, and Y407V substitutions.

66. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by T366S, T368A, and Y407V substitutions and wherein the amino acid sequence of the second

antibody constant region differs from the amino acid sequence of an IgG1 constant region by a T366W substitution.

67. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by T350V, L351Y, F405A, and Y407V substitutions and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by T350V, T366L, K392L, and T394W substitutions.

68. The protein according to claim 47, wherein the amino acid sequence of the first antibody constant region differs from the amino acid sequence of an IgG1 constant region by T350V, T366L, K392L, and T394W substitutions and wherein the amino acid sequence of the second antibody constant region differs from the amino acid sequence of an IgG1 constant region by T350V, L351Y, F405A, and Y407V substitutions.

69. The protein according to any one of claims 40-42, wherein the protein further comprises an antigen-binding site capable of binding CD16.

70. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region having the amino acid sequence of SEQ ID NO:7 and a light chain variable region having the amino acid sequence of SEQ ID NO:8.

71. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region having the amino acid sequence of SEQ ID NO:85 and a light chain variable region having the amino acid sequence of SEQ ID NO:8.

72. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region having the amino acid sequence of SEQ ID NO:1 and a light chain variable region having the amino acid sequence of SEQ ID NO:2.

73. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region

having the amino acid sequence of SEQ ID NO:3 and a light chain variable region having the amino acid sequence of SEQ ID NO:4.

74. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region
5 having the amino acid sequence of SEQ ID NO:5 and a light chain variable region having the amino acid sequence of SEQ ID NO:6.

75. A protein comprising an antigen-binding site that competes for binding to human and/or cynomolgus NKG2D with an antibody comprising a heavy chain variable region having the amino acid sequence of SEQ ID NO:9 and a light chain variable region having the
10 amino acid sequence of SEQ ID NO:20.

76. A formulation comprising the protein according to any one of claims 40-75 and a pharmaceutically acceptable carrier.

77. A cell comprising one or more nucleic acids encoding the protein according to any one of claims 40-75.

- 15 78. A method of enhancing tumor cell death, the method comprising exposing a tumor and natural killer cells to the protein according to any one of claims 40-75.

79. A method of treating cancer, wherein the method comprises administering the protein according to any one of claims 40-75 or the formulation according to claim 76 to a patient.

80. The method according to claim 79, wherein the cancer is selected from the group
20 consisting of acute myeloid leukemia, acute myelomonocytic leukemia, B cell lymphoma, bladder cancer, breast cancer, colorectal cancer, diffuse large B cell lymphoma esophageal cancer, Ewing's sarcoma, follicular lymphoma, gastric cancer, gastrointestinal cancer, gastrointestinal stromal tumors, glioblastoma, head and neck cancer, melanoma, mesothelioma, multiple myeloma, myelodysplastic syndrome, renal cell carcinoma,
25 neuroblastoma, non-small cell lung cancer, neuroendocrine tumors, ovarian cancer, and pancreatic cancer, prostate cancer, sarcomas, small cell lung cancer, T cell lymphoma, testis cancer, thymic carcinoma, thyroid cancer, urothelial cancer, cancers infiltrated by myeloid-derived suppressor cells, cancers with extracellular matrix deposition, cancers with high levels of reactive stroma, and cancers with neoangiogenesis.