CLAIMS

1. A bispecific antibody, wherein the bispecific antibody is for use in treating or preventing brain metastases from non-small cell lung cancer, wherein

the bispecific antibody comprises:

a first protein functional region targeting VEGFA, and

a second protein functional region targeting PD-1,

wherein

the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment, wherein the immunoglobulin comprises a heavy chain variable region and a light chain variable region, wherein the heavy chain variable region comprises HCDR1-HCDR3 having the amino acid sequences set forth in SEQ ID NOs: 28-30, respectively, and the light chain variable region comprises LCDR1-LCDR3 having the amino acid sequences set forth in SEQ ID NOs: 31-33, respectively; and, the single-chain variable fragment comprises a heavy chain variable region and a light chain variable region, wherein the heavy chain variable region comprises HCDR1-HCDR3 having the amino acid sequences set forth in SEQ ID NOs: 34-36, respectively, and the light chain variable region comprises LCDR1-LCDR3 having the amino acid sequences set forth in SEQ ID NOs: 37-39, respectively;

or,

the first protein functional region is a single-chain variable fragment, and the second protein functional region is an immunoglobulin, wherein the single-chain variable fragment comprises a heavy chain variable region and a light chain variable region, wherein the heavy chain

variable region comprises HCDR1-HCDR3 having the amino acid sequences set forth in SEQ ID NOs: 28-30, respectively, and the light chain variable region comprises LCDR1-LCDR3 having the amino acid sequences set forth in SEQ ID NOs: 31-33, respectively; and, the immunoglobulin comprises a heavy chain variable region and a light chain variable region, wherein the heavy chain variable region comprises HCDR1-HCDR3 having the amino acid sequences set forth in SEQ ID NOs: 34-36, respectively, and the light chain variable region of the immunoglobulin comprises LCDR1-LCDR3 having the amino acid sequences set forth in SEQ ID NOs: 37-39, respectively.

2. The bispecific antibody according to claim 1, wherein
the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment, wherein an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is selected from SEQ ID NO: 5 and SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is selected from SEQ ID NO: 7, SEQ ID NO: 11, and SEQ ID NO: 17.

3. The bispecific antibody according to any one of claims 1 to 2, wherein the bispecific antibody is selected from any one of the following (1)-(6):
(1)
an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid

sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 7;

(2)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 11;

(3)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 17;

(4)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region

of the single-chain variable fragment is set forth in SEQ ID NO: 7;

(5)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 11; and

(6)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 17.

4. The bispecific antibody according to claim 1, wherein

the first protein functional region is a single-chain variable fragment, and the second protein functional region is an immunoglobulin, wherein an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3; and, an amino acid sequence of the heavy chain variable region of the immunoglobulin is selected from SEQ ID NO: 5 and SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the

immunoglobulin is selected from SEQ ID NO: 7, SEQ ID NO: 11, and SEQ ID NO: 17.

5. The bispecific antibody according to any one of claims 1 and 4, wherein the bispecific antibody is selected from any one of the following (7)-(12):

(7)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 7; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3;

(8)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 11; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3;

(9)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 5, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 17; and, an amino acid sequence of the heavy

chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3;

(10)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 7; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3;

(11)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 11; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 3; and

(12)

an amino acid sequence of the heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 9, and an amino acid sequence of the light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 17; and, an amino acid sequence of the heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 1, and an amino acid sequence of the light chain variable

region of the single-chain variable fragment is set forth in SEQ ID NO: 3.

6. The bispecific antibody according to any one of claims 1 to 5, wherein the immunoglobulin is of a human IgG1 subtype,
wherein, according to the EU numbering system, a heavy chain constant region of the immunoglobulin comprises one of the following mutation combinations:
L234A and L235A; or
L234A, L235A, and G237A.

7. The bispecific antibody according to any one of claims 1 to 3 and 6, wherein
an amino acid sequence of the heavy chain of the immunoglobulin is set forth in SEQ ID NO: 24, and an amino acid sequence of the light chain of the immunoglobulin is set forth in SEQ ID NO: 26;
an amino acid sequence of the heavy chain of the immunoglobulin is set forth in SEQ ID NO: 20, and an amino acid sequence of the light chain of the immunoglobulin is set forth in SEQ ID NO: 26; or
an amino acid sequence of the heavy chain of the immunoglobulin is set forth in SEQ ID NO: 22, and an amino acid sequence of the light chain of the immunoglobulin is set forth in SEQ ID NO: 26.

8. The bispecific antibody according to any one of claims 1 to 7, wherein the first protein functional region and the second protein functional region are linked directly or via a linker; and/or the heavy chain variable region of the single-chain variable fragment and the light chain variable region of the single-chain variable fragment are linked directly

or via a linker.

9. The bispecific antibody according to claim 8, wherein
the linker is the polypeptide set forth in SEQ ID NO: 43 or a polypeptide formed by concatenating a plurality of (e.g., 2, 3, 4, 5, or 6) polypeptides set forth in SEQ ID NO: 43.

10. The bispecific antibody according to any one of claims 1 to 9, wherein
the first protein functional region and the second protein functional region are independently 1, 2, or more in number.

11. The bispecific antibody according to any one of claims 1 to 10, wherein
the single-chain variable fragment is two in number, and the two single-chain variable fragments are linked to the C-termini of the two heavy chains of the immunoglobulin, respectively.

12. The bispecific antibody according to any one of claims 1 to 11, wherein the single-chain variable fragment is independently a sequential linkage of: heavy chain variable region-linker-light chain variable region (VH-linker-VL), or light chain variable region-linker-heavy chain variable region (VL-linker-VH);
preferably, an amino acid sequence of the single-chain variable fragment is set forth in any one of SEQ ID NOs: 45-48.

13. The bispecific antibody according to any one of claims 1 to 12, wherein the first protein functional region is an anti-VEGFA

immunoglobulin, and the second protein functional region is an anti-PD-1 single-chain variable fragment;
preferably, two anti-PD-1 single-chain variable fragment molecules are present, and the two molecules are linked to the C-termini of the two heavy chains of the anti-VEGFA immunoglobulin, respectively;
preferably, the bispecific antibody is a bispecific antibody in the form of IgG-scFv;
preferably, the bispecific antibody is a tetravalent bispecific antibody in the form of IgG-scFv.

14. The bispecific antibody according to any one of claims 1 to 13, wherein the bispecific antibody is selected from any one of the following (13)-(16):
(13)
the bispecific antibody comprises:
a first protein functional region targeting VEGFA, and
a second protein functional region targeting PD-1;
the first protein functional region is 1 in number, and the second protein functional region is 2 in number,
wherein the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment;
an amino acid sequence of a heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of a light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3;
an amino acid sequence of a heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 9 or SEQ ID NO: 5, and an amino acid sequence of a light chain variable region of

the single-chain variable fragment is set forth in SEQ ID NO: 17, SEQ ID NO: 7, or SEQ ID NO: 11;

the single-chain variable fragment is linked to the C-termini of the two heavy chains of the immunoglobulin, respectively;

the first protein functional region and the second protein functional region are linked via a first linker; the heavy chain variable region of the single-chain variable fragment and the light chain variable region of the single-chain variable fragment are linked via a second linker; the first linker and the second linker are identical or different;

preferably, amino acid sequences of the first linker and the second linker are independently selected from SEQ ID NO: 18 and SEQ ID NO: 19;

preferably, amino acid sequences of the first linker and the second linker are each set forth in SEQ ID NO: 18;

(14)

the bispecific antibody comprises:

a first protein functional region targeting VEGFA, and

a second protein functional region targeting PD-1;

the first protein functional region is 1 in number, and the second protein functional region is 2 in number,

wherein the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment;

an amino acid sequence of a heavy chain of the immunoglobulin is set forth in SEQ ID NO: 24, SEQ ID NO: 20, or SEQ ID NO: 22, and an amino acid sequence of a light chain of the immunoglobulin is set forth in SEQ ID NO: 26;

an amino acid sequence of a heavy chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 9, and an

amino acid sequence of a light chain variable region of the single-chain variable fragment is set forth in SEQ ID NO: 17;

the single-chain variable fragment is linked to the C-termini of the two heavy chains of the immunoglobulin, respectively;

the first protein functional region and the second protein functional region are linked via a first linker; the heavy chain variable region of the single-chain variable fragment and the light chain variable region of the single-chain variable fragment are linked via a second linker; the first linker and the second linker are identical or different;

preferably, amino acid sequences of the first linker and the second linker are independently selected from SEQ ID NO: 18 and SEQ ID NO: 19;

preferably, amino acid sequences of the first linker and the second linker are each set forth in SEQ ID NO: 18;

(15)

the bispecific antibody comprises:

a first protein functional region targeting VEGFA, and

a second protein functional region targeting PD-1;

the first protein functional region is 1 in number, and the second protein functional region is 2 in number,

wherein the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment;

an amino acid sequence of a heavy chain variable region of the immunoglobulin is set forth in SEQ ID NO: 1, and an amino acid sequence of a light chain variable region of the immunoglobulin is set forth in SEQ ID NO: 3;

an amino acid sequence of the single-chain variable fragment is set forth in any one of SEQ ID NOs: 45-48;

the single-chain variable fragment is linked to the C-termini of the two heavy chains of the immunoglobulin, respectively;
the first protein functional region and the second protein functional region are linked via a first linker;
preferably, the first linker is selected from SEQ ID NO: 18 and SEQ ID NO: 19;
(16)
the bispecific antibody comprises:
a first protein functional region targeting VEGFA, and
a second protein functional region targeting PD-1;
the first protein functional region is 1 in number, and the second protein functional region is 2 in number,
wherein the first protein functional region is an immunoglobulin, and the second protein functional region is a single-chain variable fragment;
an amino acid sequence of a heavy chain of the immunoglobulin is set forth in SEQ ID NO: 24, SEQ ID NO: 20, or SEQ ID NO: 22, and an amino acid sequence of a light chain of the immunoglobulin is set forth in SEQ ID NO: 26;
an amino acid sequence of the single-chain variable fragment is set forth in any one of SEQ ID NOs: 45-48;
the single-chain variable fragment is linked to the C-termini of the two heavy chains of the immunoglobulin, respectively;
the first protein functional region and the second protein functional region are linked via a first linker;
preferably, the first linker is selected from SEQ ID NO: 18 and SEQ ID NO: 19.

15. The bispecific antibody according to any one of claims 1 to 14,

wherein the bispecific antibody is administered in combination with at least one chemotherapeutic drug.

16. The bispecific antibody according to claim 15, wherein
the chemotherapeutic drug is one or more selected from a tyrosine kinase inhibitor, a platinum-based drug (e.g., cisplatin or carboplatin), pemetrexed, paclitaxel, docetaxel, temozolomide, and fotemustine.

17. The bispecific antibody according to any one of claims 1 to 16, wherein the bispecific antibody is characterized by one or more of the following (1) to (4):
(1) a unit dose of the bispecific antibody is 100 mg-4000 mg, 200 mg-3000 mg, 200 mg-2000 mg, 300 mg-2000 mg, 400 mg-2000 mg, 500 mg-2000 mg, or 1000 mg-2000 mg;
(2) a single administration dose of the bispecific antibody is 1-50 mg, 10 mg-50 mg, 10 mg-40 mg, 10 mg-30 mg, 10 mg-20 mg, 10 mg-15 mg, 15 mg-30 mg, or 15 mg-20 mg per kg body weight;
(3) the bispecific antibody is administered once every 1 week, every 2 weeks, every 3 weeks, or every 4 weeks;
(4) the bispecific antibody is administered by intravenous drip infusion, intravenous injection, or intraperitoneal injection.

18. The bispecific antibody according to any one of claims 1 to 17, wherein
the bispecific antibody is administered in combination with pemetrexed and carboplatin; or
the bispecific antibody is administered in combination with paclitaxel and carboplatin.

19. The bispecific antibody according to claim 18, wherein the bispecific antibody is characterized by one or more of the following (1) to (3):

(1) a single administration dose of pemetrexed is 10-1500 mg/m^2, 100-1000 mg/m^2, 200-900 mg/m^2, 300-800 mg/m^2, 400-700 mg/m^2, or 500-600 mg/m^2 based on a body surface area (m^2) of each subject; preferably, pemetrexed is administered once every 1 week, every 2 weeks, every 3 weeks, or every 4 weeks;

(2) a single administration dose of carboplatin is independently AUC 2-6 min*mg/mL or AUC 2.5-5 min*mg/mL; preferably, carboplatin is administered once every 1 week, every 2 weeks, every 3 weeks, or every 4 weeks;

(3) a single administration dose of paclitaxel is 67.5-175 mg/m^2, 87.5-175 mg/m^2, or 131-175 mg/m^2 based on a body surface area (m^2) of each subject; preferably, paclitaxel is administered once every 1 week, every 2 weeks, every 3 weeks, or every 4 weeks.

20. The bispecific antibody according to any one of claims 1 to 19, wherein the brain metastases from non-small cell lung cancer are selected from one or more of brain metastases from squamous non-small cell lung cancer, brain metastases from non-squamous non-small cell lung cancer, and brain metastases from lung adenocarcinoma;

preferably, a patient with brain metastases from non-small cell lung cancer has PD-L1 TPS ≥ 50%, 1% < PD-L1 TPS ≤ 49%, or PD-L1 TPS ≤ 1%.