

Potent tumor-directed T cell activation and tumor inhibition induced by ALG.APV-527, a 4-1BB x 5T4 ADAPTIR™ bispecific antibody, 34th Annual Meeting & Pre-Conference Programs of the Society for Immunotherapy of Cancer, National Harbor, MD, 2019).

[0008] Claudin superfamily members are key components of tight junction, maintaining cellular polarity and sealing the spaces between adjacent cells. Claudin 6 (CLDN6) is a carcinoembryonic protein expressed during early development but silenced in healthy adult human tissues. CLDN6 expression has been reported in a wide range of non-hematological cancer such as pediatric brain tumors, gastric adenocarcinomas and germ cell tumors, and ovarian and testicle cancers. Its expression is often correlated with a poor prognosis. Claudin18.2 is widely expressed in a wide range of human malignancies including gastric, esophageal, pancreatic, lung, and ovarian cancers. Claudin18.2 has a superior target safety profile. In normal tissue, Claudin18.2 expression is restricted to the stomach and only on short-lived differentiated cells. Nectin family proteins mediate cell-cell adhesion through homophilic and heterophilic trans-interactions, in which heterophilic trans-interactions are much stronger than homophilic trans-interaction. As a Nectin protein family member, nectin-4 is an important driver for tumorigenesis and metastasis. Over-expression of Nectin-4 in cancer tissue is associated with cancer progression and poor prognosis.

[0009] Cancers of epithelial origin represent a significant global medical challenge that impacts patients, their families, and the health care system. For the unmet medical needs of patients with tumors overexpress epithelial tumor antigens, such as CLDN6, CLDN18.2, or Nectin-4, specific binding proteins that bind CD137 and these tumor antigens that alone, or in combination with other agents can be used for antibody-based immunotherapy, providing potentially effective and safe treatment solutions.

SUMMARY

[0010] The present disclosure addresses the above need by providing bispecific binding proteins that bind to CD137 and a tumor associated antigen (TAA). In particular examples the disclosure provides bispecific antibodies that bind to the tumor specific antigens CLDN18.2, CLDN6 or Nectin-4, and to the costimulatory CD137 receptor. Such bispecific binding proteins may be useful for the treatment of a disease or disorder such as cancer.

[0011] Also provided herein is a bispecific binding protein that binds a tumor associated antigen and CD137 comprising: (a) an antibody scaffold module comprising a first antigen-binding site that binds the tumor associated antigen and a second antigen-binding site that binds the tumor associated antigen; and (b) at least one first binding module comprising a third antigen-binding site that binds CD137.

[0012] In some examples, the tumor associated antigen is selected from the group consisting of: Claudin 6, Claudin 18.2, and Nectin-4.

[0013] In some examples, the tumor associated antigen is Claudin 6.

[0014] In some examples, the tumor associated antigen is Claudin 18.2.

[0015] In some examples, the tumor associated antigen is Nectin-4.

[0016] In some examples, the antibody scaffold module is an IgG.

[0017] In some examples, the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise: (i) a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 45, CDR2: SEQ ID NO: 46, and CDR3: SEQ ID NO: 47; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 48, CDR2: SEQ ID NO: 49, and CDR3: SEQ ID NO: 50; or (ii) a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 51, CDR2: SEQ ID NO: 52, and CDR3: SEQ ID NO: 53; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 54, CDR2: SEQ ID NO: 55, and CDR3: SEQ ID NO: 56.

[0018] In some examples, the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 25 or SEQ ID NO: 27; and a light chain variable region sequence as set forth in SEQ ID NO: 26 or SEQ ID NO: 28.

[0019] In some examples, the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise: (i) a heavy chain variable region sequence as set forth in SEQ ID NO: 25 and a light chain variable region sequence as set forth in SEQ ID NO: 26; or (ii) a heavy chain variable region sequence as set forth in SEQ ID NO: 27 and a light chain variable region sequence as set forth in SEQ ID NO: 28.

[0020] In some examples, the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise: a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 33, CDR2: SEQ ID NO: 34, and CDR3: SEQ ID NO: 35; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 36, CDR2: SEQ ID NO: 37, and CDR3: SEQ ID NO: 38.

[0021] In some examples, the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21; and a light chain variable region sequence as set forth in SEQ ID NO: 22.

[0022] In some examples, the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise: a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 57, CDR2: SEQ ID NO: 58, and CDR3: SEQ ID NO: 59; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 60, CDR2: SEQ ID NO: 61, and CDR3: SEQ ID NO: 62.

[0023] In some examples, the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 29 or SEQ ID NO: 31; and a light chain variable region sequence as set forth in SEQ ID NO: 30 or SEQ ID NO: 32.

[0024] In some examples, the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise: (i) a heavy chain variable region sequence as set forth in SEQ ID NO: 29 and a light chain variable region sequence as set forth in SEQ ID NO: 30; or (ii) a heavy chain variable region sequence as set forth in SEQ ID NO: 31 and a light chain variable region sequence as set forth in SEQ ID NO: 32.

[0025] In some examples, the bispecific binding protein comprises one first binding module.

[0026] In some examples, the binding protein comprises two first binding modules.

[0027] In some examples, the first binding module is an antibody fragment.

[0028] In some examples, the antibody fragment is an scFv.

[0029] In some examples, the first binding module binds CD137.

[0030] In some examples, the first binding module is an scFV that binds CD137.

[0031] In some examples, the antibody scaffold module comprises two heavy chain sequences both having a C-terminus and an N-terminus, and wherein the antibody scaffold module comprises two light chain sequences both having a C-terminus and a N-terminus. The first binding module is covalently attached to the C-terminus of one or both of the antibody scaffold module heavy chain sequences, the C-terminus of one or both of the antibody scaffold module light chain sequences, the N-terminus of one or both of the antibody scaffold module heavy chain sequences, the N-terminus of one or both of the antibody scaffold module light chain sequences, or combinations thereof, and wherein the first binding module and the antibody scaffold module are covalently attached to each other directly or through an interlinker.

[0032] In some examples, the first binding module and the antibody scaffold module are covalently attached to each other through an interlinker having a sequence as set forth in SEQ ID NO: 64 or SEQ ID NO: 65.

[0033] In some examples, the first binding module is covalently attached to the C-terminus of both of the antibody scaffold module heavy chain sequences.

[0034] In some examples, the first binding module is covalently attached to the C-terminus of both of the antibody scaffold module light chain sequences.

[0035] In some examples, the first binding module is covalently attached to the N-terminus of both of the antibody scaffold module heavy chain sequences.

[0036] In some examples, the first binding module binds CD137 and comprises: a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 39, CDR2: SEQ ID NO: 40, and CDR3: SEQ ID NO: 41; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 42, CDR2: SEQ ID NO: 43, and CDR3: SEQ ID NO: 44.

[0037] In some examples, the first binding module bind CD137 and comprises: a heavy chain variable region sequence as set forth in SEQ ID NO: 23; and a light chain variable region sequence as set forth in SEQ ID NO: 24.

[0038] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21, and a light chain variable region sequence as set forth in SEQ ID NO: 22; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

[0039] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0040] In some examples, the heavy chain variable region sequence and the light chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0041] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0042] In some examples, the heavy chain of the antibody scaffold module, the glycine-serine linker, and the first binding module comprise a sequence as set forth in SEQ ID NO: 3.

[0043] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21, and a light chain variable region sequence as set forth in SEQ ID NO: 22; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the C-terminus of each light chain in the antibody scaffold module by a glycine-serine linker.

[0044] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0045] In some examples, the heavy chain variable region sequence and the light chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0046] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0047] In some examples, the light chain of the antibody scaffold module, the glycine-serine linker, and the first binding module comprise a sequence as set forth in SEQ ID NO: 5.

[0048] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 25 or 27, and a light chain variable region sequence as set forth in SEQ ID NOs: 26 or 28; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

[0049] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0050] In some examples, the heavy chain variable region sequence and the heavy chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0051] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0052] In some examples, the heavy chain of the antibody scaffold module, the glycine-serine linker, and the first binding module comprise a sequence as set forth in SEQ ID NO: 12, 13 or 72.

[0053] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the C-terminus of each light chain in the antibody scaffold module by a glycine-serine linker.

[0054] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0055] In some examples, the heavy chain variable region sequence and the light chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0056] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0057] In some examples, the light chain of the antibody scaffold module, the glycine-serine linker, and the first binding module comprise a sequence as set forth in SEQ ID NO: 17.

[0058] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the N-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

[0059] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0060] In some examples, the heavy chain variable region sequence and the light chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0061] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0062] In some examples, the first binding module, the glycine-serine linker, and the heavy chain of the antibody scaffold module comprise a sequence as set forth in SEQ ID NO: 18.

[0063] In some examples, the bispecific binding protein comprises two first binding modules that bind CD137 and wherein: the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32; the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24, wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

[0064] In some examples, the glycine-serine linker is a 3xG4S linker (SEQ ID NO: 64).

[0065] In some examples, the heavy chain variable region sequence and the light chain variable region sequence in the first binding modules are attached by a glycine-serine linker.

[0066] In some examples, the glycine-serine linker is a 4xG4S linker (SEQ ID NO: 65).

[0067] In some examples, the heavy chain of the antibody scaffold module, the glycine-serine linker, and the first binding module comprise a sequence as set forth in SEQ ID NO: 14.

[0068] In some examples, the antibody scaffold module further comprises a constant region.

[0069] In some examples, the constant region comprises one or more Fc silencing mutations.

[0070] In some examples, the Fc silencing mutation can be L234A/L235A (LALA) alone or in combination with P329A mutation (LALAP) or N297A

[0071] In some examples, the constant region comprises SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68, SEQ ID NO: 69, or SEQ ID NO: 73.

[0072] Also provided herein is a bispecific binding protein that binds a tumor associated antigen and CD137 comprising: (a) an antibody scaffold module comprising a means for binding the tumor associated antigen via a first antigen-binding site and a second antigen-binding site; and (b) at least one first binding module comprising a means for binding CD137 via a third antigen-binding site.

[0073] The present disclosure also provides a pharmaceutical composition comprising the bispecific binding protein as disclosed herein and a pharmaceutically acceptable carrier.

[0074] The present disclosure also provides a method of treating or preventing cancer, the method comprising administering the bispecific binding protein as disclosed herein to a patient in need thereof.

[0075] Also provided herein is an isolated polynucleotide comprising a sequence encoding the bispecific binding protein as disclosed herein. The present disclosure also provides vectors or cells comprising the polynucleotide disclosed herein. Also provided herein is a method for the production of the bispecific binding protein as disclosed herein comprising culturing the cell disclosed herein.

[0075a] In one aspect, the present disclosure relates to bispecific binding protein that binds a tumor associated antigen and CD137 comprising: (a) an antibody scaffold module comprising a first antigen-binding site that binds the tumor associated antigen and a second antigen-binding site that binds the tumor associated antigen; (b) at least one first binding module comprising a third antigen-binding site that binds CD137; wherein the tumor associated antigen is Claudin 6, Claudin 18.2, or Nectin 4.

[0075b] In another aspect, the present disclosure relates to a pharmaceutical composition comprising the bispecific binding protein as disclosed herein and a pharmaceutically acceptable carrier.

[0075c] In another aspect, the present disclosure relates to use of the bispecific binding as disclosed herein in the manufacture of a medicament for treating or preventing cancer.

[0075d] In another aspect, the present disclosure relates to an isolated polynucleotide comprising a sequence encoding the bispecific binding protein as disclosed herein.

[0075e] In another aspect, the present disclosure relates to a vector comprising the polynucleotide as disclosed herein.

[0075f] In another aspect, the present disclosure relates to a cell comprising a polynucleotide as disclosed herein, and/or a vector as disclosed herein.

[0075g] In another aspect, the present disclosure relates to a method for the production of the bispecific binding protein as disclosed herein, the method comprising culturing the cell as disclosed herein.

WHAT IS CLAIMED IS:

1. A bispecific binding protein that binds a tumor associated antigen and CD137 comprising:
 - (a) an antibody scaffold module comprising a first antigen-binding site that binds the tumor associated antigen and a second antigen-binding site that binds the tumor associated antigen;
 - (b) at least one first binding module comprising a third antigen-binding site that binds CD137;wherein the tumor associate antigen is Claudin 6, Claudin 18.2, or Nectin 4.
2. The bispecific binding protein of claim 1, wherein the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise:
 - (i) a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 45, CDR2: SEQ ID NO: 46, and CDR3: SEQ ID NO: 47; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 48, CDR2: SEQ ID NO: 49, and CDR3: SEQ ID NO: 50; or
 - (ii) a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 51, CDR2: SEQ ID NO: 52, and CDR3: SEQ ID NO: 53; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 54, CDR2: SEQ ID NO: 55, and CDR3: SEQ ID NO: 56.
3. The bispecific binding protein of claim 2, wherein the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise:
 - (i) a heavy chain variable region sequence as set forth in SEQ ID NO: 25 and a light chain variable region sequence as set forth in SEQ ID NO: 26; or
 - (ii) a heavy chain variable region sequence as set forth in SEQ ID NO: 27 and a light chain variable region sequence as set forth in SEQ ID NO: 28.
4. The bispecific binding protein of claim 1, wherein the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise:
 - a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 33, CDR2: SEQ ID NO: 34, and CDR3: SEQ ID NO: 35; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 36, CDR2: SEQ ID NO: 37, and CDR3: SEQ ID NO: 38.

- 5.** The bispecific binding protein of claim **4**, wherein the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21; and a light chain variable region sequence as set forth in SEQ ID NO: 22.
- 6.** The bispecific binding protein of claim **1**, wherein the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise:
a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 57, CDR2: SEQ ID NO: 58, and CDR3: SEQ ID NO: 59; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 60, CDR2: SEQ ID NO: 61, and CDR3: SEQ ID NO: 62.
- 7.** The bispecific binding protein of claim **6**, wherein the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise:
(i) a heavy chain variable region sequence as set forth in SEQ ID NO: 29 and a light chain variable region sequence as set forth in SEQ ID NO: 30; or
(ii) a heavy chain variable region sequence as set forth in SEQ ID NO: 31 and a light chain variable region sequence as set forth in SEQ ID NO: 32.
- 8.** The bispecific binding protein of any one of claims **1-7**, wherein the first binding module is an antibody fragment.
- 9.** The bispecific binding protein of any one of claims **1-8**, wherein the first binding module is an scFV that binds CD137.
- 10.** The bispecific binding protein of any one of claims **1-9**, wherein the first binding module and the antibody scaffold module are covalently attached to each other through an interlinker having a sequence as set forth in SEQ ID NO: 64 or SEQ ID NO: 65.

11. The bispecific binding protein of any one of claims **1-10**, wherein the first binding module binds CD137 and comprises:

a heavy chain variable region sequence comprising CDR1: SEQ ID NO: 39, CDR2: SEQ ID NO: 40, and CDR3: SEQ ID NO: 41; and a light chain variable region sequence comprising CDR1: SEQ ID NO: 42, CDR2: SEQ ID NO: 43, and CDR3: SEQ ID NO: 44.

12. The bispecific binding protein of claim **11**, wherein the first binding module bind CD137 and comprises: a heavy chain variable region sequence as set forth in SEQ ID NO: 23; and a light chain variable region sequence as set forth in SEQ ID NO: 24.

13. The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:

the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21, and a light chain variable region sequence as set forth in SEQ ID NO: 22;

the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,

wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

14. The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:

the first antigen-binding site and the second antigen-binding site both bind Claudin 18.2 and comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 21, and a light chain variable region sequence as set forth in SEQ ID NO: 22;

the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,

wherein the first binding modules are separately attached to the C-terminus of each light chain in the antibody scaffold module by a glycine-serine linker.

15. The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:

the first antigen-binding site and the second antigen-binding site both bind Claudin 6 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 25 or 27, and a light chain variable region sequence as set forth in SEQ ID NOs: 26 or 28;

the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,

wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

16. The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:

the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32;

the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,

wherein the first binding modules are separately attached to the C-terminus of each light chain in the antibody scaffold module by a glycine-serine linker.

17. The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:

the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32;

the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,

wherein the first binding modules are separately attached to the N-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.

- 18.** The bispecific binding protein of any one of claims **1-12**, wherein the bispecific binding protein comprises two first binding modules that bind CD137 and wherein:
- the first antigen-binding site and the second antigen-binding site both bind Nectin-4 and comprise a heavy chain variable region sequence as set forth in SEQ ID NOs: 29 or 31, and a light chain variable region sequence as set forth in SEQ ID NOs: 30 or 32;
- the first binding modules each comprise a heavy chain variable region sequence as set forth in SEQ ID NO: 23, and a light chain variable region sequence as set forth in SEQ ID NO: 24,
- wherein the first binding modules are separately attached to the C-terminus of each heavy chain in the antibody scaffold module by a glycine-serine linker.
- 19.** The bispecific binding protein of any one of claims **1-18**, wherein the antibody scaffold module further comprises a constant region.
- 20.** The binding protein of claim **19**, wherein the constant region comprises SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68, SEQ ID NO: 69 or SEQ ID NO: 73.
- 21.** A pharmaceutical composition comprising the bispecific binding protein of any one of claims **1-20** and a pharmaceutically acceptable carrier.
- 22.** Use of the bispecific binding protein of any one of claims **1-20** in the manufacture of a medicament for treating or preventing cancer.
- 23.** An isolated polynucleotide comprising a sequence encoding the bispecific binding protein of any one of claims **1-20**.
- 24.** A vector comprising the polynucleotide of claim **23**.
- 25.** A cell comprising a polynucleotide of claim **23**, and/or a vector of claim **24**.

26. A method for the production of the bispecific binding protein of any one of claims **1-20**, the method comprising culturing the cell of claim **25**.