

In another aspect, the present disclosure refers to a tetra-specific antibody, comprising the tetra-specific antibody monomer as described herein, wherein the tetra-specific antibody comprises an amino acid sequence having a percentage homology to SEQ ID NO. 38 or 40, wherein the percentage homology is not less than 98%. In yet another aspect, the present disclosure refers to a tetra-specific antibody, comprising the tetra-specific antibody monomer as described herein, wherein the tetra-specific monomer comprises: (i) 3 complementarity determining regions (CDRs) of SEQ ID No. 26 and 3 CDRs of SEQ ID No. 28; (ii) 3 CDRs of SEQ ID No. 2 and 3 CDRs of SEQ ID No. 4; (iii) 3 CDRs of SEQ ID No. 14 and 3 CDRs of SEQ ID No. 16; and/or (iv) 3 CDRs of SEQ ID No. 10 and 3 CDRs of SEQ ID No. 12. In yet another aspect, the present disclosure refers to an isolated nucleic acid sequence, encoding the tetra-specific antibody as described herein comprising an amino acid sequence having a percentage homology to SEQ ID NO. 38 or 40, wherein the percentage homology is not less than 98%.

In yet another aspect, the present disclosure refers to an expression vector, comprising the isolated nucleic acid sequence as described herein. In yet another aspect, the present disclosure refers to a host cell comprising the isolated nucleic acid sequence as described herein, wherein the host cell is a prokaryotic cell or a eukaryotic cell. In a further aspect, the present disclosure refers to a method for producing a tetra-specific antibody, comprising culturing a host cell as described herein comprising an isolated nucleic acid sequence such that the DNA sequence encoding the tetra-specific antibody is expressed, and purifying said tetra-specific antibody, wherein the isolated nucleic acid sequence encodes an amino acid having a percentage homology to SEQ ID NO. 37-40, wherein the percentage homology is not less than 98%.

In a further aspect, the present disclosure refers to a pharmaceutical composition, comprising a pharmaceutically acceptable carrier and the tetraspecific antibody as described herein. In a further aspect, the present disclosure refers to the use of the tetra-specific antibody as described herein in the manufacture of a medicament for treating a human subject with a cancer, wherein an effective amount of the tetra-specific antibody is to be administered to the subject. In yet another aspect, the present disclosure refers to the use of the tetra-specific antibody as described herein and an effective amount of a therapeutic agent in the manufacture of a combination therapy for treating a human subject with a cancer, wherein an effective amount of the therapeutic agent is to be co-administered, and wherein the therapeutic agent comprises an antibody, a chemotherapy agent, an enzyme, an anti-estrogen agent, a receptor tyrosine kinase inhibitor, a kinase inhibitor, a cell cycle inhibitor, a check point inhibitor, a DNA, RNA or protein synthesis inhibitor, a RAS inhibitor, an inhibitor of PD1, PD-L1, CTLA4, 4-1BB, OX40, GITR, ICOS, LIGHT, TIM3, LAG3, TIGIT, CD40, CD27, HVEM, BTLA, VISTA, B7H4, CSF1R, NKG2D, CD73, CD3, CD19 or a combination thereof.

In one example, the application provides tetra-specific antibody monomers. In one example, the tetra-specific antibody monomer has a N-terminal and a C-terminal and include in tandem from the N-terminal to the C-terminal, a first scFv domain at the N-terminal, a second scFv domain, a Fab domain, a Fc domain, and a third scFv at the C-terminal. The first scFv domain, the Fab domain, the second scFv domain, and the third scFv domain each has a binding specificity against a different antigen.

In one example, the antigen includes a tumor antigen, an immune signaling antigen, or a combination thereof. In one example, the first scFv domain, the Fab domain, the second scFv domain, and the third scFv domain each has a binding specificity against a tumor antigen or an immune signaling antigen. In one example, the first scFv domain has a binding specificity against a tumor antigen. In one example, the first scFv domain has a binding specificity against an immune signaling antigen. In one example, the second scFv domain has a binding specificity against a tumor antigen. In one example, the second scFv domain has a binding specificity against an immune signaling antigen. In one example, the Fab domain has a binding specificity against a tumor antigen. In one example, the Fab domain has a binding

CLAIMS

What is claimed is:

1. A tetra-specific antibody monomer having a N-terminal and a C-terminal, comprising in tandem from the N-terminal to the C-terminal,

a first scFv domain at the N-terminal,

a second scFv domain

a Fab domain,

a Fc domain, and

a third scFv at the C-terminal,

wherein the first scFv domain, the second scFv domain, the Fab domain, and the third scFv domain each has a binding specificity against a different antigen, and wherein the antigen is a tumor antigen, an immune signaling antigen, or a combination thereof.

2. The tetra-specific antibody monomer of Claim 1, wherein the first scFv domain, the second scFv domain, the Fab domain, and the third scFv domain each independently has a binding specificity against an antigen selected from ROR1, PD-L1, CD3, CD28, CEA, HER2, EGFRvIII, EGFR, LMP1, LMP2A, Mesothelin, PSMA, EpCAM, glypimay-3, gpA33, GD2, TROP2, NKG2D, BCMA, CD19, CD20, CD33, CD123, CD22, CD30, PD1, OX40, 4-1BB, GITR, TIGIT, TIM-3, LAG-3, CTLA4, CD40, VISTA, ICOS, BTLA, LIGHT, HVEM, CSF1R, CD73, and CD39, and wherein the Fc domain comprises a human IgG Fc domain.

3. The tetra-specific antibody monomer of Claim 1, wherein the first scFv domain, the second scFv domain, the Fab domain, and the third scFv domain each independently has a binding specificity against an antigen selected from CD19, CD3, CD137, 4-1BB, and PD-L1.

4. The tetra-specific antibody monomer of Claim 1, wherein the first scFv domain has a binding specificity against CD19.

5. The tetra-specific antibody monomer of Claim1, wherein the second scFv domain has a binding specificity against CD3.

6. The tetra-specific antibody monomer of Claim 1, wherein the Fab domain has a binding specificity against 4-1BB or CD137.

7. The tetra-specific antibody monomer of Claim1, wherein the third scFv domain has a binding specificity against PD-L1.

8. The tetra-specific antibody monomer of Claim 1, wherein the first scFv domain has a binding specificity against CD19, wherein the second scFv domain has a binding specificity against CD3, wherein

the Fab domain has a binding specificity against 4-1BB or CD137, and wherein the third scFv domain has a binding specificity against PD-L1.

9. The tetra-specific antibody monomer of Claim 1, wherein the first scFv domain, the second scFv domain, or the third scFv domain comprises a gly-gly-gly-gly-ser (G4S)_n linker, wherein n is 2, 3 or 4.

10. The tetra-specific antibody monomer of Claim 1, comprising an amino acid sequence having a percentage homology to SEQ ID No. 38 or 40, wherein the percentage homology is not less than 98%.

11. The scFv domain of the tetra-specific antibody monomer of Claim 1, comprising an amino acid sequence having a percentage homology to SEQ ID No. 2, 4, 6, 8, 10, 12, 26, 28, 30 or 32, wherein the percentage homology is not less than 98%.

12. The Fab domain of the tetra-specific antibody monomer of Claim 1, comprising an amino acid sequence having a percentage homology to SEQ ID NO. 2, 4, 6, 8, 10, 12, 26, 28, 30 or 32, wherein the percentage homology is not less than 90%.

13. The tetra-specific antibody monomer of Claim 1, wherein the monomer comprises: (i) 3 complementarity determining regions (CDRs) of SEQ ID No. 26 and 3 CDRs of SEQ ID No. 28; (ii) 3 CDRs of SEQ ID No. 2 and 3 CDRs of SEQ ID No. 4; (iii) 3 CDRs of SEQ ID No. 14 and 3 CDRs of SEQ ID No. 16; and/or (iv) 3 CDRs of SEQ ID No. 10 and 3 CDRs of SEQ ID No. 12.

14. The tetra-specific antibody monomer of Claim 13, wherein the monomer comprises: (i) 3 complementarity determining regions (CDRs) of SEQ ID No. 26 and 3 CDRs of SEQ ID No. 28; (ii) 3 CDRs of SEQ ID No. 2 and 3 CDRs of SEQ ID No. 4; (iii) 3 CDRs of SEQ ID No. 14 and 3 CDRs of SEQ ID No. 16; and (iv) 3 CDRs of SEQ ID No. 10 and 3 CDRs of SEQ ID No. 12.

15. A tetra-specific antibody, comprising the tetra-specific antibody monomer of Claim 13 or 14.

16. A tetra-specific antibody, comprising the tetra-specific antibody monomer of Claim 1, wherein the tetra-specific antibody comprises an amino acid sequence having a percentage homology to SEQ ID NO. 38 or 40, wherein the percentage homology is not less than 98%.

17. An isolated nucleic acid sequence, encoding the tetra-specific antibody of Claim 16 comprising an amino acid sequence having a percentage homology to SEQ ID NO. 38 or 40, wherein the percentage homology is not less than 98%.

18. An expression vector, comprising the isolated nucleic acid sequence of Claim 17.

19. A host cell comprising the isolated nucleic acid sequence of Claim 17, wherein the host cell is a prokaryotic cell or a eukaryotic cell.

20. A method for producing a tetra-specific antibody, comprising culturing a host cell of Claim 19 comprising an isolated nucleic acid sequence such that the DNA sequence encoding the tetra-specific antibody is expressed, and purifying said tetra-specific antibody, wherein the isolated nucleic acid

sequence encodes an amino acid having a percentage homology to SEQ ID NO. 38 or 40, wherein the percentage homology is not less than 98%.

21. A pharmaceutical composition, comprising a pharmaceutically acceptable carrier and the tetra-specific antibody of Claim 15 or 16.

22. Use of the tetra-specific antibody according to Claim 15 or 16 in the manufacture of a medicament for treating a human subject with a cancer, wherein an effective amount of the tetra-specific antibody is to be administered to the subject.

23. Use of the tetra-specific antibody according to Claim 15 or 16 and an effective amount of a therapeutic agent in the manufacture of a combination therapy for treating a human subject with a cancer, wherein an effective amount of the therapeutic agent is to be co-administered, and wherein the therapeutic agent comprises an antibody, a chemotherapy agent, an enzyme, an anti-estrogen agent, a receptor tyrosine kinase inhibitor, a kinase inhibitor, a cell cycle inhibitor, a check point inhibitor, a DNA, RNA or protein synthesis inhibitor, a RAS inhibitor, an inhibitor of PD1, PD-L1, CTLA4, 4-1BB, OX40, GITR, ICOS, LIGHT, TIM3, LAG3, TIGIT, CD40, CD27, HVEM, BTLA, VISTA, B7H4, CSF1R, NKG2D, CD73, CD3, CD19 or a combination thereof.