Claims

1. An antibody that specifically recognizes glypican-3 (GPC3), wherein the antibody comprises a heavy chain variable region and a light chain variable region, wherein the heavy chain variable region comprises: CDR1 of the amino acid sequence of SEQ ID NO: 73, CDR2 of the amino acid sequence of SEQ ID NO: 74 and CDR3 of the amino acid sequence of SEQ ID NO: 75; and the light chain variable region comprises CDR1 of the amino acid sequence of SEQ ID NO: 76, CDR2 of the amino acid sequence of SEQ ID NO: 77 and CDR3 of the amino acid sequence of SEQ ID NO: 78.

2. The antibody of claim 1, wherein the antibody is a humanized monoclonal antibody comprising a heavy chain variable region and a light chain variable region, wherein the heavy chain variable region thereof comprises: CDR1 of the amino acid sequence of SEQ ID NO: 73, CDR2 of the amino acid sequence of SEQ ID NO: 74 and CDR3 of the amino acid sequence of SEQ ID NO: 75; and the light chain variable region thereof comprises CDR1 of the amino acid sequence of SEQ ID NO: 76, CDR2 of the amino acid sequence of SEQ ID NO: 77 and CDR3 of the amino acid sequence of SEQ ID NO: 78.

3. The antibody of claim 2, wherein the antibody has a heavy chain variable region of the amino acid sequence of SEQ ID NO: 58 and a light chain variable region of the amino acid sequence of SEQ ID NO: 59.

4. The antibody of claim 1, wherein the antibody has a heavy chain variable region of the amino acid sequence of SEQ ID NO: 81 and a light chain variable region of the amino acid sequence of SEQ ID NO: 82.

5. A nucleic acid encoding the antibody of any one of claims 1-4.

6. An expression vector comprising the nucleic acid of claim 5.

7. A host cell comprising the expression vector of claim 6 or having the nucleic acid of claim 5 integrated into its genome.

8. *In vitro* use of the antibody of any one of claims 1-4 for the preparation of a targeted

drug, antibody-drug conjugate, or a polyfunctional antibody that specifically targets tumor cells expressing glypican-3; or

for the preparation of a reagent for diagnosing a tumor expressing Glypican-3; or

for the preparation of chimeric antigen receptor-modified immune cells.

9. A multi-functional immunoconjugate, comprising:

the antibody of any one of claims 1-4; and

a functional molecule linked thereto; and the functional molecule is selected from a group consisting of a molecule targeting a tumor surface marker, a tumor-suppressing molecule, a molecule targeting a surface marker of an immune cell, or a detectable label.

10. The multifunctional immunoconjugate of claim 9, wherein the molecule that targets a tumor surface marker is an antibody or ligand that binds to a tumor surface marker; or

the tumor-suppressing molecule is an anti-tumor cytokine or anti-tumor toxin; and preferably, the cytokine includes: IL-12, IL-15, IFN-beta and TNF-alpha.

11. The multifunctional immunoconjugate of claim 9, wherein the detectable label includes a fluorescent label or a chromogenic label.

12. The multifunctional immunoconjugate of claim 10, wherein the antibody that binds to a tumor surface marker refers to an antibody that recognizes antigens other than glypican-3, wherein the other antigens include: EGFR, EGFRvIII, mesothelin, HER2, EphA2, Her3, EpCAM, MUC1, MUC16, CEA, Claudin 18.2, folate receptor, Claudin 6, WT1, NY-ESO- 1, MAGE 3, ASGPR1 or CDH16.

13. The multifunctional immunoconjugate of claim 9, wherein the molecule that targets the surface marker of the immune cell is an antibody that binds to T cell surface marker, which can form a T-cell-engaging bifunctional antibody with the antibody of any one of claims 1-4.

14. The multifunctional immunoconjugate of claim 13, wherein the antibody that binds to the immune cell surface marker is an anti-CD3 antibody.

15. The multifunctional immunoconjugate of claim 14, wherein the multifunctional immunoconjugate is a fusion peptide, and further comprises a linker peptide between the antibody of any one of claims 1-4 and the functional molecule linked thereto.

16. A nucleic acid encoding the multi-functional immunoconjugate of any one of claims 9-15.

17. *In vitro* use of the multifunctional immunoconjugate of any one of claims 9-15, for the preparation of an antineoplastic agent, or

for the preparation of an agent for diagnosis of tumors that express Glypican-3; or

for the preparation of chimeric antigen receptor-modified immune cells; preferably, the immune cells include T lymphocyte, NK cell or NKT lymphocyte.

18. A chimeric antigen receptor comprising the antibody of any one of claims 1-4, a transmembrane region and an intracellular signal region, which are sequentially linked.

19. The chimeric antigen receptor of claim 18, wherein the intracellular signal region is selected from a group consisting of intracellular signal region sequences of CD3ζ, FcεRIγ, CD27, CD28, CD137, CD134, MyD88, CD40 or a combination thereof.

20. The chimeric antigen receptor of claim 18, wherein the transmembrane region comprises a transmembrane region of CD8 or CD28.

21. The chimeric antigen receptor of claim 18, wherein the chimeric antigen receptor comprises the following sequentially connected antibody, a transmembrane region and an intracellular signal region:

The antibody of any one of claims 1-4, CD8, and CD3ζ;

The antibody of any one of claims 1-4, CD8, CD137 and CD3ζ;

The antibody of any one of claims 1-4, the transmembrane region of CD28 molecule, the intracellular signaling region of CD28 molecule and CD3ζ; or

The antibody of any one of claims 1-4, the transmembrane region of CD28 molecule, the intracellular signaling region of CD28 molecule, CD137 and CD3ζ.

22. The chimeric antigen receptor of claim 18, wherein the antibody is a single chain

antibody or a single domain antibody.

23. The chimeric antigen receptor of claim 18, wherein the chimeric antigen receptor comprises:

The amino acid sequence of SEQ ID NO: 86;

The amino acid sequence of SEQ ID NO: 88; or

The amino acid sequence of SEQ ID NO: 90.

24. A nucleic acid encoding the chimeric antigen receptor of any one of claims 18-23.

25. An expression vector comprising the nucleic acid of claim 24.

26. A virus comprising the vector of claim 25.

27. *In vitro* use of the chimeric antigen receptor of any one of claims 18-23, or the nucleic acid of claim 24, or the expression vector of claim 25 or the virus of claim 26 for the preparation of genetically modified immune cells targeting the tumor that expresses Glypican-3.

28. The use of claim 27, wherein the tumor expressing glypican-3 includes liver cancer, melanoma, ovarian clear cell carcinoma, yolk sac tumor and neuroblastoma.

29. A genetically modified immune cell, which has the chimeric antigen receptor of any one of claims 18-23 expressed on its surface.

30. The immune cell of claim 29, wherein it further carries an exogenous encoding sequence for cytokines; and preferably, the cytokines include IL-12, IL-15 or IL-21.

31. The immune cell of claim 29, wherein it also expresses another chimeric antigen receptor which does not contain CD3ζ but contains the intracellular signaling domain of CD28, the intracellular signaling domain of CD137, or a combination of both.

32. The immune cell of claim 29, wherein it further expresses a chemokine receptor;

and preferably, the chemokine receptor includes: CCR2.

33. The immune cell of claim 29, wherein it further expresses siRNA which can reduce expression of PD-1 or a protein which blocks PD-L1.

34. The immune cell of claim 29, wherein it further expresses a safety switch; and preferably, the safety switch includes iCaspase-9, Truancated EGFR or RQR8.

35. The immune cell of claim 29, wherein the immune cells include T lymphocytes, NK cells or NKT cells.

36. *In vitro* use of the genetically modified immune cell of any one of claims 29-35 for the preparation of a tumor-inhibiting drug, and the tumor is a tumor expressing glypican-3.

37. A pharmaceutical composition comprising:

The antibody of any one of claims 1-4 or a nucleic acid encoding the antibody; or

The immunoconjugate of any one of claims 9-15; or

The chimeric antigen receptor of any one of claims 18-23; or

The genetically modified immune cell of any one of claims 29-35.