

## CLAIMS

1. An isolated monoclonal antibody or antigen-binding fragment thereof comprising a heavy chain complementarity determining region 1 (HCDR1), HCDR2, HCDR3, a light chain complementarity determining region 1 (LCDR1), LCDR2, and LCDR3, having the polypeptide sequences of SEQ ID NOs: 87, 88, 89, 90, 91 and 92, respectively, or SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively; wherein the antibody or antigen-binding fragment thereof specifically binds GPC3.
2. The isolated monoclonal antibody or antigen-binding fragment thereof of claim 1, comprising a heavy chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 85, 99-102, or 183-197, or a light chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 86, 103-105, or 198-201.
3. The isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-2, comprising:
  - a. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:85, and a light chain variable region having the polypeptide sequence of SEQ ID NO:86;
  - b. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:99, and a light chain variable region having the polypeptide sequence of SEQ ID NO:103;
  - c. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:99, and a light chain variable region having the polypeptide sequence of SEQ ID NO:104;
  - d. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:100, and a light chain variable region having the polypeptide sequence of SEQ ID NO:103;
  - e. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:100, and a light chain variable region having the polypeptide sequence of SEQ ID NO:104;
  - f. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:101, and a light chain variable region having the polypeptide sequence of SEQ ID NO:103;

- g. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:101,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:104;
- h. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:102,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:105;
- i. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 100,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
198;
- j. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 183,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- k. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 183,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
198;
- l. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 185,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- m. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- n. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 185,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- o. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- p. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 189,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;

- q. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 188,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
103;
- r. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 190,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- s. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- t. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- u. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- v. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- w. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 189,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- x. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 190,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- y. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- z. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;

- aa. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- bb. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- cc. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 189,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- dd. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 190,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ee. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ff. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- gg. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- hh. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ii. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- jj. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;

- kk. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
  - ll. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
  - mm. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:  
185, and a light chain variable region having the polypeptide sequence of SEQ ID  
NO: 200;
  - nn. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
  - oo. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
  - pp. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
  - qq. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 196,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
201; or
  - rr. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 197,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
201.
4. The isolated monoclonal antibody or antigen-binding fragment thereof of any one of  
claims 1-3, wherein the antibody or antigen-binding fragment thereof is chimeric and/or human  
or humanized.
  5. The isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-  
4, wherein the antibody or antigen-binding fragment thereof is capable of inducing effector-  
mediated tumor cell lysis through antibody-dependent cellular cytotoxicity (ADCC), antibody-  
dependent cellular phagocytosis (ADCP), and/or complement-dependent cytotoxicity (CDC),

and/or mediating the recruitment of conjugated drugs, and/or forming a bispecific antibody with another mAb or antigen-binding fragment thereof with cancer-killing effect.

6. A bispecific antibody or antigen-binding fragment thereof comprising the monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-5.

7. An isolated nucleic acid encoding the monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-5 or the bispecific antibody or antigen-binding fragment thereof of claim 6.

8. A vector comprising the isolated nucleic acid of claim 7.

9. A host cell comprising the vector of claim 8.

10. A pharmaceutical composition, comprising the isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-5 or the bispecific antibody or antigen-binding fragment thereof of claim 6 and a pharmaceutically acceptable carrier.

11. Use of the pharmaceutical composition of claim 10 in the manufacture of a medicament for the treatment of a GPC3 expressing cancer and/ or an inflammatory disease in a subject in need thereof, wherein the medicament is to be administered to the subject.

12. The use of claim 11, wherein the GPC3 expressing cancer is selected from the group consisting of a lung cancer, a gastric cancer, an esophageal cancer, a bile duct cancer, a cholangiocarcinoma, a colon cancer, a hepatocellular carcinoma, a renal cell carcinoma, a bladder urothelial carcinoma, a metastatic melanoma, a breast cancer, an ovarian cancer, a cervical cancer, a head and neck cancer, a pancreatic cancer, a glioma, a glioblastoma, and other solid tumors, and a non-Hodgkin's lymphoma (NHL), an acute lymphocytic leukemia (ALL), a chronic lymphocytic leukemia (CLL), a chronic myelogenous leukemia (CML), a multiple myeloma (MM), an acute myeloid leukemia (AML), and other liquid tumors.

13. The pharmaceutical composition of claim 10 for use in therapy.

14. A method of producing the monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-5 or the bispecific antibody or antigen-binding fragment thereof of claim 6, comprising culturing a cell comprising a nucleic acid encoding the monoclonal antibody or antigen-binding fragment thereof or bispecific antibody or antigen-binding fragment thereof under conditions to produce the monoclonal antibody or antigen-binding fragment thereof or bispecific antibody or antigen-binding fragment thereof and recovering the monoclonal antibody

or antigen-binding fragment thereof or bispecific antibody or antigen-binding fragment thereof from the cell or culture.

15. A method of producing a pharmaceutical composition comprising the monoclonal antibody or antigen-binding fragment of any one of claims 1-5 or the bispecific antibody or antigen-binding fragment thereof of claim 6, comprising combining the monoclonal antibody or antigen-binding fragment thereof or bispecific antibody or antigen-binding fragment thereof with a pharmaceutically acceptable carrier to obtain the pharmaceutical composition.

16. A method of determining the level of a GPC3 in a subject, the method comprising:

- a. being provided with a sample from the subject;
- b. contacting the sample with an isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-5; and
- c. determining the level of a GPC3 in the subject,

optionally wherein the sample is a tissue sample or a blood sample, optionally, wherein the tissue sample is a cancer tissue sample.

17. An isolated polynucleotide comprising a nucleic acid sequence encoding a chimeric antigen receptor (CAR), wherein the CAR comprises:

- a. an extracellular domain comprising at least one antigen binding domain that specifically binds GPC3;
- b. a hinge region;
- c. a transmembrane region; and
- d. an intracellular signaling domain.

18. The isolated polynucleotide of claim 17, wherein the antigen binding domain comprises a heavy chain complementarity determining region 1 (HCDR1), HCDR2, HCDR3, a light chain complementarity determining region 1 (LCDR1), LCDR2, and LCDR3, having the polypeptide sequences of SEQ ID NOs: 87, 88, 89, 90, 91 and 92, respectively, or SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively.

19. The isolated polynucleotide of claim 17 or 18, wherein the antigen binding domain comprises a heavy chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 85, 99-102, or 183-197, or a light chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 86, 103-105, or 198-201.

20. The isolated polynucleotide of claim 19, wherein the antigen binding domain comprises:

- a. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:85,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:86;
- b. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:99,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:103;
- c. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:99,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:104;
- d. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:100,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:103;
- e. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:100,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:104;
- f. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:101,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:103;
- g. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:101,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:104;
- h. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:102,  
and a light chain variable region having the polypeptide sequence of SEQ ID  
NO:105;
- i. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 100,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
198;
- j. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 183,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;

- k. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 183,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
198;
- l. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 185,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- m. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- n. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 185,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- o. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- p. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 189,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- q. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 188,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
103;
- r. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 190,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- s. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- t. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;

- u. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
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- v. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
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and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
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and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
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- y. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- z. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- aa. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- bb. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- cc. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 189,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- dd. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 190,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;

- ee. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 191,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ff. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 192,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- gg. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 193,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- hh. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 186,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ii. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;
- jj. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
199;
- kk. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 195,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- ll. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 194,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
200;
- mm. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:  
185, and a light chain variable region having the polypeptide sequence of SEQ ID  
NO: 200;
- nn. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187,  
and a light chain variable region having the polypeptide sequence of SEQ ID NO:  
104;

- oo. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 200;
  - pp. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 187, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 199;
  - qq. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 196, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 201; or
  - rr. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 197, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 201.
21. The isolated polynucleotide of any one of claims 17-20, wherein the antigen binding domain is a single chain variable fragment (scFv) that specifically binds GPC3, preferably human GPC3, optionally wherein the scFv is humanized.
22. The isolated polynucleotide of claim 21, wherein the single chain variable fragment (scFv) comprises a polypeptide sequence at least 95% identical to any one of SEQ ID NOs: 167-170.
23. The isolated polynucleotide of any one of claims 17-22, wherein the chimeric antigen receptor (CAR) comprises one or more antigen binding domains, and/or wherein the intracellular signaling domain comprises one or more costimulatory domains and one or more activating domains.
24. A chimeric antigen receptor (CAR) encoded by the isolated polynucleotide of any one of claims 17-23.
25. A vector comprising the isolated polynucleotide of any one of claims 17-23.
26. A host cell comprising the vector of claim 25, wherein the host cell is a T cell or NK cell, preferably a human T cell or NK cell.
27. A method of making a host cell expressing a chimeric antigen receptor (CAR), the method comprising transducing a T cell or NK cell with the vector of claim 25.
28. A method of producing a chimeric antigen receptor (CAR)-T cell or CAR-NK cell, the method comprising culturing T cells or NK cells comprising the isolated polynucleotide

comprising a nucleic acid encoding a chimeric antigen receptor (CAR) of any one of claims 17-23 under conditions to produce the CAR-T cell and recovering the CAR-T cell or CAR-NK cell.

29. A method of generating a cell comprising a chimeric antigen receptor (CAR), the method comprising contacting a cell with the isolated polynucleotide comprising a nucleic acid encoding a chimeric antigen receptor (CAR) of any one of claims 17-23, wherein the isolated polynucleotide is an *in vitro* transcribed RNA or synthetic RNA.

30. Use of the host cell of claim 26 in the manufacture of a medicament for the treatment of a GPC3 expressing cancer in a subject in need thereof, wherein the medicament is to be administered to the subject.

31. The use of claim 30, wherein the GPC3 expressing cancer is selected from a lung cancer, a gastric cancer, a colon cancer, a hepatocellular carcinoma, a renal cell carcinoma, a bladder urothelial carcinoma, a metastatic melanoma, a breast cancer, an ovarian cancer, a cervical cancer, a head and neck cancer, a pancreatic cancer, a glioma, a glioblastoma, and other solid tumors, and a non-Hodgkin's lymphoma (NHL), an acute lymphocytic leukemia (ALL), a chronic lymphocytic leukemia (CLL), a chronic myelogenous leukemia (CML), a multiple myeloma (MM), an acute myeloid leukemia (AML), and other liquid tumors.

32. The use of claim 30 or claim 31, further comprising an agent that increases the efficacy of a cell expressing a CAR, an agent that ameliorates one or more side effects associated with administration of a cell expressing a CAR, and/or an anti-GPC3 monoclonal antibody or bispecific antibody to be administered to the subject in need thereof.

33. The host cell of claim 26 for use in therapy.