

CLAIMS

~~It is claimed:—~~

~~1.~~

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:

- (1) SEQ ID NOs: 87, 88, 89, 90, 91 and 92, respectively; or SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively;
- (2) SEQ ID NOs: 108, 109, 110, 111, 112 and 113, respectively; or SEQ ID NOs: 114, 115, 116, 117, 118 and 119, respectively;
- (3) SEQ ID NOs: 17, 18, 19, 20, 21, and 22, respectively; or SEQ ID NOs: 23, 24, 25, 26, 27 and 28, respectively;
- (4) SEQ ID NOs: 31, 32, 33, 34, 35, and 36, respectively; or SEQ ID NOs: 37, 38, 39, 40, 41 and 42, respectively;
- (5) SEQ ID NOs: 45, 46, 47, 48, 49, and 50, respectively; or SEQ ID NOs: 51, 52, 53, 54, 55 and 56, respectively;
- (6) SEQ ID NOs: 59, 60, 61, 62, 63, and 64, respectively; ~~or~~ or SEQ ID NOs: 65, 66, 67, 68, 69 and 70, respectively; or
- (7) SEQ ID NOs: 73, 74, 75, 76, 77, and 78, respectively; or SEQ ID NOs: 79, 80, 81, 82, 83 and 84, respectively;

wherein the antibody or antigen-binding fragment thereof specifically binds GPC3, preferably human GPC3.

~~2.~~ ~~2.~~—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:

- ~~(1) SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively;~~
- ~~(2) SEQ ID NOs: 114, 115, 116, 117, 118 and 119, respectively;~~
- ~~(3) SEQ ID NOs: 23, 24, 25, 26, 27, and 28, respectively;~~
- ~~(4) SEQ ID NOs: 37, 38, 39, 40, 41, and 42, respectively;~~

(5) SEQ ID NOs: 51, 52, 53, 54, 55, and 56, respectively;

(6) SEQ ID NOs: 65, 66, 67, 68, 69, and 70, respectively; or

(7) SEQ ID NOs: 79, 80, 81, 82, 83, and 84, respectively;

wherein the antibody or antigen-binding fragment thereof specifically binds GPC3, preferably human GPC3.

3. 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:

- (1) SEQ ID NOs: 3, 4, 5, 6, 7, and 8, respectively; or SEQ ID NOs: 9, 10, 11, 12, 13 and 14, respectively;

wherein the antibody or antigen-binding fragment thereof specifically binds TIP-1, preferably human TIP-1.

4. 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:

- (1) SEQ ID NOs: 9, 10, 11, 12, 13, and 14, respectively;

wherein the antibody or antigen-binding fragment thereof specifically binds TIP-1, preferably human TIP-1.

3. 5.-The isolated monoclonal antibody or antigen-binding fragment thereof of ~~any one of claims 1-4~~claim 1 or 3, comprising a heavy chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 85, 106, 15, 29, 43, 57, 71, 1, 99-102, 120-123, 126-132, 139-140, 143-149, 153-156, 160-163, 183-197, or 202-205, or a light chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 86, 107, 16, 30, 44, 58, 72, ~~or 22, 103-105, 124-125, 133-138, 141-142, 150-152, 157-159, 164-166, or 198-201.~~

4. 6.-The isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-~~5~~3, 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:106, and a light chain variable region having the polypeptide sequence of SEQ ID NO:107;
- c. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:15, and a light chain variable region having the polypeptide sequence of SEQ ID NO:16;
- d. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:29, and a light chain variable region having the polypeptide sequence of SEQ ID NO:30;
- e. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:43, and a light chain variable region having the polypeptide sequence of SEQ ID NO:44;
- f. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:57, and a light chain variable region having the polypeptide sequence of SEQ ID NO:58;
- g. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:71, and a light chain variable region having the polypeptide sequence of SEQ ID NO:72;
or
- h. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:1, and a light chain variable region having the polypeptide sequence of SEQ ID NO:2;

~~7. The isolated monoclonal antibody or antigen binding fragment thereof of any one of claims 1-6, wherein the antibody or antigen binding fragment thereof is chimeric and/or human or humanized.~~

~~8. The isolated monoclonal antibody or antigen binding fragment thereof of claim 7, wherein the humanized monoclonal antibody or antigen binding fragment thereof comprises a heavy chain variable region having a polypeptide sequence at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identical to any one of SEQ ID NOs: 99-102, 120-123, 126-132, 139-140, 143-149, 153-156, 160-163, 183-197, or 202-205, or a light chain variable region having a polypeptide sequence at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identical to any one of SEQ ID NOs: 103-105, 124-125, 133-138, 141-142, 150-152, 157-159, 164-166, or 198-201.~~

~~9. The isolated monoclonal antibody or antigen binding fragment thereof of claim 8, wherein the humanized monoclonal antibody or antigen binding fragment thereof comprises:~~

- i. 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;
- j. 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;
- k. 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;
- l. 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;
- m. 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;
- n. 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;
- o. 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;
- p. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:120,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:124;
- q. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:120,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:125;
- r. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:121,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:124;

- s. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:121,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:125;
- t. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:122,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:124;
- u. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:122,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:125;
- v. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:123,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:124;
- w. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:139,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:141;
- x. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:139,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:142;
- y. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:140,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:141;
- z. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:140,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:142;
- aa. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:153,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:157;
- bb. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:153,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:158;

- cc. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:154,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:157;
- dd. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:154,
and a light chain variable region having the polypeptide sequence of SEQ ID
NO:158;
- ee. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 143,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
150;
- ff. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 144,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
150;
- gg. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 145,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
150;
- hh. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 147,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
151;
- ii. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 147,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
152;
- jj. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 148,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
151;
- kk. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 148,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
152;
- ll. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 149,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
151;

- mm. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 149, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 152;
- nn. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 160, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- oo. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 161, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- pp. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 162, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- qq. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 163, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- rr. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 205, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- ss. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 202, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- tt. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 203, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- uu. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 204, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 164;
- vv. 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;

- ww. 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;
- xx. 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;
- yy. 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;
- zz. 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;
- aaa. 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;
- bbb. 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;
- ccc. 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;
- ddd. 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;
- eee. 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;
- fff. 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;

- ggg. 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;
- hhh. 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;
- iii. 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;
- jjj. 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;
- kkk. 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;
- lll. 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;
- mmm. 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;
- nnn. 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;
- ooo. 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;
- ppp. 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;

- qqq. 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;
- rrr. 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;
- sss. 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;
- ttt. 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;
- uuu. 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;
- vvv. 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;
- www. 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;
- xxx. 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;
- yyy. 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;
- zzz. 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;

- aaaa. 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;
- bbbb. 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;
- cccc. 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;
- dddd. 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
- eeee. 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.

5. ~~10.~~ The isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-~~94~~, wherein the antibody or antigen-binding fragment thereof is chimeric and/or human or humanized.

6. The isolated monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-~~5~~, 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.

~~4~~7. A bispecific antibody or antigen-binding fragment thereof comprising the monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-~~106~~.

~~4~~8. An isolated nucleic acid encoding the monoclonal antibody or antigen-binding fragment thereof of any one of claims 1-~~106~~ or the bispecific antibody or antigen-binding fragment thereof of claim 7.

~~9. ~~13.~~ An isolated nucleic acid encoding the bispecific antibody or antigen-binding fragment thereof of claim 11.~~

~~14.~~ A vector comprising the isolated nucleic acid of claim ~~12 or 13~~8.

~~15.~~10. A host cell comprising the vector of claim ~~14~~9.

~~16.~~11. A pharmaceutical composition, comprising the isolated monoclonal antibody or antigen-binding fragment thereof of any one of ~~claims 1-10 or 6~~or the bispecific antibody or antigen-binding fragment thereof of claim ~~14~~7 and a pharmaceutically acceptable carrier.

~~17. A method of targeting~~12. Use of the pharmaceutical composition of claim 11 in the manufacture of a TAA or medicament for the treatment of a cancer cell surface, and/or treating a cancer, and/or treating or an inflammatory disease in a subject in need thereof, ~~comprising administering~~ wherein the medicament is to be administered to the subject ~~the pharmaceutical composition of claim 16,~~ optionally wherein the 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.~~13. ~~18~~The pharmaceutical composition of claim 11 for use in therapy.

14. A method of producing the monoclonal antibody or antigen-binding fragment thereof of any one of claims ~~1-10 or 6~~or the bispecific antibody or antigen-binding fragment thereof of claim ~~14~~7, 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.

~~19~~15. A method of producing a pharmaceutical composition comprising the monoclonal antibody or antigen-binding fragment of any one of claims ~~1-10 or 6~~or the bispecific antibody or antigen-binding fragment thereof of claim ~~14~~7, 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.

~~20~~16. A method of determining the level of a TAA in a subject, the method comprising:

- a. ~~obtaining~~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-~~10~~6; and
- c. determining the level of a TAA in the subject₂.

~~21. The method of claim 20, optionally~~ wherein the sample is a tissue sample or a blood sample, optionally₂ wherein the tissue sample is a cancer tissue sample.

~~22~~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.

~~23~~18. The isolated polynucleotide of claim ~~22~~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:

- (1) SEQ ID NOs: 87, 88, 89, 90, 91 and 92, respectively₂, or SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively;
- (~~2~~) SEQ ID NOs: 108, 109, 110, 111, 112 and 113, respectively;
- (~~3~~) SEQ ID NOs: 17, 18, 19, 20, 21, and 22, respectively;
- (~~4~~) SEQ ID NOs: 31, 32, 33, 34, 35, and 36, respectively;
- (~~5~~) SEQ ID NOs: 45, 46, 47, 48, 49, and 50, respectively;
- (~~6~~) SEQ ID NOs: 59, 60, 61, 62, 63, and 64, respectively; or
- (~~7~~) SEQ ID NOs: 73, 74, 75, 76, 77, and 78, respectively;

~~wherein the antigen binding domain specifically binds GPC3, preferably human GPC3.~~

~~24. The isolated polynucleotide of claim 22, 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:~~

- (1) ~~SEQ ID NOs: 93, 94, 95, 96, 97 and 98, respectively;~~
~~(8)(2)~~ , or SEQ ID NOs: 114, 115, 116, 117, 118 and 119, respectively;
~~(9)(3)~~ SEQ ID NOs: 17, 18, 19, 20, 21, and 22, respectively, or SEQ ID NOs: 23, 24, 25, 26, 27, and 28, respectively;
~~(10)(4)~~ SEQ ID NOs: 31, 32, 33, 34, 35, and 36, respectively, or SEQ ID NOs: 37, 38, 39, 40, 41, and 42, respectively;
~~(11)(5)~~ SEQ ID NOs: 45, 46, 47, 48, 49, and 50, respectively, or SEQ ID NOs: 51, 52, 53, 54, 55, and 56, respectively; ~~or~~
~~(12)(6)~~ SEQ ID NOs: 59, 60, 61, 62, 63, and 64, respectively, or SEQ ID NOs: 65, 66, 67, 68, 69, and 70, respectively; ~~or~~
~~(13)(7)~~ SEQ ID NOs: 73, 74, 75, 76, 77, and 78, respectively, or SEQ ID NOs: 79, 80, 81, 82, 83, and 84, respectively;

~~wherein the antigen binding domain specifically binds GPC3, preferably human GPC3.~~

~~2519.~~ The isolated polynucleotide of ~~any one of claims 22-24~~ 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, 106, 15, 29, 43, 57, 71, 99-102, 120-123, 126-132, 139-140, 143-149, 153-156, 160-163, 183-197, or 71202-205, or a light chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 86, 107, 16, 30, 44, 58, ~~or 7272~~, 103-105, 124-125, 133-138, 141-142, 150-152, 157-159, 164-166, or 198-201.

~~2620.~~ The isolated polynucleotide of claim ~~2519~~, 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:106, and a light chain variable region having the polypeptide sequence of SEQ ID NO:107;
- c. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:15, and a light chain variable region having the polypeptide sequence of SEQ ID NO:16;
- d. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:29, and a light chain variable region having the polypeptide sequence of SEQ ID NO:30;
- e. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:43, and a light chain variable region having the polypeptide sequence of SEQ ID NO:44;

- f. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:57,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:58;

or

- g. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:71,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:72;

~~27. The isolated polynucleotide of any one of claims 22-24, wherein the antigen binding domain is humanized and comprises a heavy chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 99-102, 120-123, 126-132, 139-140, 143-149, 153-156, 160-163, 183-197, or 202-205, or a light chain variable region having a polypeptide sequence at least 95% identical to SEQ ID NO: 103-105, 124-125, 133-138, 141-142, 150-152, 157-159, 164-166, or 198-201.~~

~~28. The isolated polynucleotide of claim 27, wherein the antigen binding domain is humanized and comprises:~~

- h. 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;
- i. 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;
- j. 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;
- k. 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;
- l. 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;
- m. 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;

- n. 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;
- o. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:120, and a light chain variable region having the polypeptide sequence of SEQ ID NO:124;
- p. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:120, and a light chain variable region having the polypeptide sequence of SEQ ID NO:125;
- q. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:121, and a light chain variable region having the polypeptide sequence of SEQ ID NO:124;
- r. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:121, and a light chain variable region having the polypeptide sequence of SEQ ID NO:125;
- s. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:122, and a light chain variable region having the polypeptide sequence of SEQ ID NO:124;
- t. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:122, and a light chain variable region having the polypeptide sequence of SEQ ID NO:125;
- u. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:123, and a light chain variable region having the polypeptide sequence of SEQ ID NO:124;
- v. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:139, and a light chain variable region having the polypeptide sequence of SEQ ID NO:141;
- w. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:139, and a light chain variable region having the polypeptide sequence of SEQ ID NO:142;

- x. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:140, and a light chain variable region having the polypeptide sequence of SEQ ID NO:141;
- y. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:140, and a light chain variable region having the polypeptide sequence of SEQ ID NO:142;
- z. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:153, and a light chain variable region having the polypeptide sequence of SEQ ID NO:157;
- aa. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:153, and a light chain variable region having the polypeptide sequence of SEQ ID NO:158;
- bb. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:154, and a light chain variable region having the polypeptide sequence of SEQ ID NO:157;
- cc. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:154, and a light chain variable region having the polypeptide sequence of SEQ ID NO:158;
- dd. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 143, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 150;
- ee. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 144, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 150;
- ff. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 145, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 150;
- gg. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 147, and a light chain variable region having the polypeptide sequence of SEQ ID NO: 151;

- hh. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 147,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
152;
- ii. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 148,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
151;
- jj. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 148,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
152;
- kk. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 149,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
151;
- ll. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 149,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
152;
- mm. a heavy chain variable region having the polypeptide sequence of SEQ ID NO:
160, and a light chain variable region having the polypeptide sequence of SEQ ID
NO: 164;
- nn. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 161,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- oo. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 162,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- pp. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 163,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- qq. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 205,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;

- rr. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 202,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- ss. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 203,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- tt. a heavy chain variable region having the polypeptide sequence of SEQ ID NO: 204,
and a light chain variable region having the polypeptide sequence of SEQ ID NO:
164;
- uu. 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;
- vv. 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;
- ww. 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;
- xx. 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;
- yy. 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;
- zz. 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;
- aaa. 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;

- bbb. 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;
- ccc. 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;
- ddd. 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;
- eee. 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;
- fff. 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;
- ggg. 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;
- hhh. 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;
- iii. 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;
- jjj. 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;
- kkk. 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;

- lll. 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;
- mmm. 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;
- nnn. 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;
- ooo. 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;
- ppp. 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;
- qqq. 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;
- rrr. 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;
- sss. 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;
- ttt. 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;
- uuu. 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;

- vvv. 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;
- www. 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;
- xxx. 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;
- yyy. 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;
- zzz. 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;
- aaaa. 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;
- bbbb. 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;
- cccc. 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
- dddd. 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.

2921. The isolated polynucleotide of any one of claims ~~22-28~~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.

~~3022.~~ The isolated polynucleotide of claim 29, ~~wherein the single chain variable fragment (scFv) is humanized.~~

~~31.~~ The isolated polynucleotide of claim 29 or ~~3021~~, wherein the single chain variable fragment (scFv) comprises a polypeptide sequence at least 95% identical to any one of SEQ ID NOs: 167-182.

~~3223.~~ The isolated polynucleotide of any one of claims ~~17-22-34~~, wherein the chimeric antigen receptor (CAR) comprises one or more antigen binding domains.

~~33.~~ The isolated polynucleotide of any one of claims ~~22-32~~, and/or wherein the intracellular signaling domain comprises one or more costimulatory domains and one or more activating domains.

~~3424.~~ A chimeric antigen receptor (CAR) encoded by the isolated polynucleotide of any one of claims ~~22-33~~ 17-23.

~~3525.~~ A vector comprising the isolated polynucleotide of any one of claims ~~22-33~~ 17-23.

~~3626.~~ A host cell comprising the vector of claim ~~35~~.

~~37.~~ The host cell of claim ~~3625~~, wherein the host cell is a T cell, ~~preferably a human T cell.~~

~~38.~~ The host cell of claim ~~36~~, wherein the host cell is a T or NK cell, preferably a human T cell or NK cell.

~~3927.~~ 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 ~~3525~~.

~~4028.~~ A method of producing a chimeric antigen receptor (CAR)-T cell or CAR-NK cell, the method comprising culturing T cells ~~comprising the isolated polynucleotide comprising a nucleic acid encoding a chimeric antigen receptor (CAR) of any one of claims 22-33 under conditions to produce the CAR-T cell and recovering the CAR-T cell.~~

~~41.~~ A method of making a host cell expressing a chimeric antigen receptor (CAR), the method comprising transducing a NK cell with a vector of claim ~~35~~.

~~42.~~ A method of producing a chimeric antigen receptor (CAR)-NK cell, the method comprising culturing ~~or~~ NK cells comprising the isolated polynucleotide comprising a nucleic acid encoding a chimeric antigen receptor (CAR) of any one of claims ~~22-33~~ 17-23 under conditions to produce the CAR-~~NK~~ T cell and recovering the CAR-T cell or CAR-NK cell.

~~4329.~~ 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 ~~22-33~~17-23, wherein the isolated polynucleotide is an *in vitro* transcribed RNA or synthetic RNA.

~~44. A method~~30. Use of treating the host cell of claim 26 or 27 in the manufacture of a medicament for the treatment of cancer in a subject in need thereof, ~~comprising administering~~wherein the medicament is to be administered to the subject ~~in need thereof~~the ~~host cell of any one of claims 36-38~~, optionally wherein the 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.

~~45~~31. The method ~~use of claim 44~~30, further comprising ~~administering to the subject in need thereof~~ an agent that increases the efficacy of a cell expressing a CAR;

~~46. The method of claim 44, further comprising administering to the subject in need thereof, an agent that ameliorates one or more side effects associated with administration of a cell expressing a CAR, and/or an agent that treats the disease associated with GPC3 to be~~administered to the subject in need thereof.

~~47. The method of claim 44, further comprising administering to the subject in need thereof an agent that treats the disease associated with GPC3.~~

32. The host cell of claim 26 or 27 for use in therapy.