CLAIMS

1. A polypeptide complex, comprising a TIGIT-binding moiety and a PVRIG-binding moiety, wherein:

the TIGIT-binding moiety comprises a heavy chain variable region (VH) comprising a heavy chain CDR (HCDR) 1, a HCDR2, and a HCDR3, and a light chain variable region (VL) comprising a light chain CDR (LCDR) 1, a LCDR2 and a LCDR3, wherein:

the HCDR1 comprises the amino acid sequence of SEQ ID NO: 1 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 1, the HCDR2 comprises the amino acid sequence of SEQ ID NO: 2 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 2, the HCDR3 comprises the amino acid sequence of SEQ ID NO: 3 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 3, the LCDR1 comprises the amino acid sequence of SEQ ID NO: 4 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 4, the LCDR2 comprises the amino acid sequence of SEQ ID NO: 5 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 5, and the LCDR3 comprises the amino acid sequence of SEQ ID NO: 6 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 6;

wherein the substitution is a conservative substitution.

2. The polypeptide complex of claim 1, wherein the PVRIG-binding moiety comprises a VH comprising a HCDR1, a HCDR2, and a HCDR3, and a VL comprising a LCDR1, a LCDR2 and a LCDR3, wherein:

the HCDR1 comprises the amino acid sequence of SEQ ID NO: 7 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 7, the HCDR2 comprises the amino acid sequence of SEQ ID NO: 8 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 8, the HCDR3 comprises the amino acid sequence of SEQ ID NO: 9 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 9, the LCDR1 comprises the amino acid sequence of SEQ ID NO: 10 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 10, the LCDR2 comprises the amino acid sequence of SEQ ID NO: 11 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 11, and the LCDR3 comprises the amino acid sequence of SEQ ID NO: 12 or an amino acid sequence with substitution of no more than 1, 2 or 3 amino acids compared with SEQ ID NO: 12, wherein the substitution is a conservative substitution.

3. The polypeptide complex of claim 1 or 2, wherein the TIGIT-binding moiety and PVRIG-binding moiety are in any of a Fab and a scFv format.

4. The polypeptide complex of claim 1 or 2, wherein the HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3 of the TIGIT-binding moiety comprise the amino acid sequences of SEQ ID NOs: 1-6, respectively; and the HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3 of the PVRIG-binding moiety comprise the amino acid sequences of SEQ ID NOs: 7-12, respectively; optionally, the VH of the TIGIT-binding moiety comprises the amino acid sequence of SEQ ID NO: 13 or an amino acid sequence at least 85%, 90%, or 95% identical to SEQ ID NO: 13, and/or the VL of the TIGIT-binding moiety comprises the amino acid sequence of SEQ ID NO: 14 or an amino acid sequence at least 85%, 90%, or 95% identical to SEQ ID NO: 14; optionally, the VH of the PVRIG-binding moiety comprises the amino acid sequence of SEQ ID NO: 15 or an amino acid sequence at least 85%, 90%, or 95% identical to SEQ ID NO: 15, and/or
the VL of the PVRIG-binding moiety comprises the amino acid sequence of SEQ ID NO: 16 or an amino acid sequence at least 85%, 90%, or 95% identical to SEQ ID NO: 16.

5. The polypeptide complex of any of the preceding claims, wherein:
the TIGIT-binding moiety is in Fab format and the PVRIG-binding moiety is in scFv format;
the TIGIT-binding moiety is in scFv format and the PVRIG-binding moiety is in Fab format; or
both the TIGIT-binding moiety and the PVRIG-binding moiety are in Fab format.

6. The polypeptide complex of any of the preceding claims, wherein the polypeptide complex further comprises an immunoglobulin constant region, such as an IgG constant region, such as a human IgG1, IgG4, IgG2, IgG3 Fc region or a variant thereof; optionally, the human IgG Fc region is a human IgG1 Fc region or a variant thereof; optionally, the variant comprises one or more substitutions to modulate receptor binding or effector function, to promote dimerization, to prevent glycosylation, and/or to extend its half-life.

7. The polypeptide complex of claim 6, wherein the polypeptide complex comprises:
(a) the TIGIT-binding moiety operably linked to the Fc region and the Fc region operably linked to the PVRIG-binding moiety;
(b) the TIGIT-binding moiety operably linked to the PVRIG-binding moiety and the PVRIG-binding moiety operably linked to the Fc region; or
(c) the PVRIG-binding moiety operably linked to the TIGIT-binding moiety and the TIGIT-binding moiety operably linked to the Fc region.

8. The polypeptide complex of any of the preceding claims, comprising one, two or more TIGIT-binding moieties, as well as one, two or more PVRIG-binding moieties; optionally, the TIGIT-binding moieties are identical or different, and/or the PVRIG-binding moieties are identical or different.

9. The polypeptide complex of any of the preceding claims, comprising two heavy chains and two light chains, wherein the TIGIT-binding moiety is in Fab format and the PVRIG-binding moiety is in scFv format, and wherein from N-terminus to C-terminus:
the first and second heavy chains each comprises domains that are operably linked into the following format: VH1-CH1-Fc-scFv or scFv-VH1-CH1-Fc,
the first and second light chains each comprises domains that are operably linked into the following format: VL1-CL,
wherein the VH1-CH1 and VL1-CL are from the TIGIT-binding moiety and the scFv is from the PVRIG-binding moiety; optionally, the polypeptide complex comprises two heavy chains and two light chains, wherein the TIGIT-binding moiety is in scFv format and the PVRIG-binding moiety is in Fab format, and wherein from N-terminus to C-terminus:
the first and second heavy chains each comprises domains that are operably linked into the following format: scFv-VH2-CH1-Fc or VH2-CH1-Fc-scFv,
the first and second light chains each comprises domains that are operably linked into the following format: VL2-CL,
wherein the scFv is from the TIGIT-binding moiety and the VH2-CH1 and VL2-CL are from the PVRIG-binding moiety; optionally, the polypeptide complex comprises two heavy chains and four light chains, wherein the TIGIT-binding moiety and the PVRIG-binding moiety are in Fab format, and wherein from N-terminus to C-terminus:
the first and second heavy chains each comprises domains that are operably linked into the following format: VH1-CH1-Fc-VH2-CH1, VH2-CH1-Fc-VH1-CH1, VH1-CH1-VH2-CH1-Fc or VH2-CH1-VH1-CH1-Fc,
the first and second light chains each comprises domains that are operably linked into the following format: VL1-CL,
the third and fourth light chains each comprises domains that are operably linked into the following format: VL2-CL,
wherein the VH1-CH1 and VL1-CL are from the TIGIT-binding moiety and the VH2-CH1 and VL2-CL are from the PVRIG-binding moiety; optionally, the polypeptide complex comprises two heavy chains and four light chains, wherein the TIGIT-binding moiety and the PVRIG-binding moiety are in Fab format, and wherein from N-terminus to C-terminus:
the first and second heavy chains each comprises domains that are operably linked into the following format: VH1-C1-Fc-VH2-CH1, VH2-CH1-Fc-VH1-C1, VH1-C1-VH2-CH1-Fc or VH2-CH1-VH1-C1-Fc,
the first and second light chains each comprises domains that are operably linked into the following

format: VL1-C2,
the third and fourth light chains each comprises domains that are operably linked into the following format: VL2-CL,
wherein the VH1-C1 and VL1-C2 are from the TIGIT-binding moiety and the VH2-CH1 and VL2-CL are from the PVRIG-binding moiety; optionally,

the polypeptide complex comprises two heavy chains and four light chains, wherein the TIGIT-binding moiety and the PVRIG-binding moiety are in Fab format, and wherein from N-terminus to C-terminus:
the first and second heavy chains each comprises domains that are operably linked into the following format: VH1-CH1-Fc-VH2-C1, VH2-C1-Fc-VH1-CH1, VH1-CH1-VH2-C1-Fc or VH2-C1-VH1-CH1-Fc,
the first and second light chains each comprises domains that are operably linked into the following format: VL1-CL,
the third and fourth light chains each comprises domains that are operably linked into the following format: VL2-C2,
wherein the VH1-CH1 and VL1-CL are from the TIGIT-binding moiety and the VH2-C1 and VL2-C2 are from the PVRIG-binding moiety.

10. The polypeptide complex of any of claims 7-9, wherein operably linked is via a direct linkage or via a peptide linker; optionally, the peptide linker is a hinge region or a GS linker, such as (GS)n, (GGS)n, (GGGS)n, (GGGGS)n, (GGSG)n, (GGGSS)n, wherein n is an integer of 1-9.

11. The polypeptide complex of claim 9, comprising:
(i) a first and a second heavy chain that comprise SEQ ID NO: 17, and a first and a second light chain that comprise SEQ ID NO: 18; or
(ii) a first and a second heavy chain that comprise SEQ ID NO: 19, and a first and a second light chain that comprise SEQ ID NO: 20.

12. An isolated nucleic acid molecule, comprising a nucleic acid sequence encoding the heavy chain variable region and/or the light chain variable region of the polypeptide complex of any of claims 1-11.

13. A vector comprising the isolated nucleic acid molecule of claim 12.

14. A host cell comprising the vector of claim 13 or the isolated nucleic acid molecule of claim 12.

15. A pharmaceutical composition comprising the polypeptide complex of any of claims 1-11 and a pharmaceutically acceptable carrier.

16. A method for producing the polypeptide complex of any of claims 1-11 comprising the steps of:

- culturing a host cell comprising an expression vector(s) encoding the polypeptide complex under suitable conditions; and

- harvesting the polypeptide complex from the cell culture.

17. A method for treating or preventing a cancer or an immune related disorder in a subject (such as human and non-human mammals such as mouse), comprising administering an effective amount of the polypeptide complex of any of claims 1-11 or the pharmaceutical composition of claim 15 to the subject; optionally, the cancer is selected from colon cancer, lung cancer, breast cancer, ovarian cancer, melanoma, bladder cancer, renal cell carcinoma, liver cancer, prostate cancer, stomach cancer, pancreatic cancer, lymphoma, leukemia, uterine cancer, cervical cancer, testicular cancer, esophageal cancer, gastrointestinal cancer, gastric cancer, colorectal cancer, kidney cancer, clear cell renal carcinoma, head and neck cancer, germ cell cancer, bone cancer, thyroid cancer, skin cancer, neoplasm of the central nervous system, mesothelioma, myeloma, and sarcoma, for example the cancer is colon cancer; optionally, the immune related disorder is a T cell dysfunctional disorder or an infection.

18. The method of claim 17, wherein the method further comprises administering an additional therapeutic agent to the subject.

19. Use of the polypeptide complex of any of claims 1-11 in the manufacture of a medicament for treating or preventing cancer or a T cell dysfunctional disorder or an infection; optionally, the cancer is selected from colon cancer, lung cancer, breast cancer, ovarian cancer, melanoma, bladder cancer, renal cell carcinoma, liver cancer, prostate cancer, stomach cancer, pancreatic cancer, lymphoma, leukemia, uterine cancer, cervical cancer, testicular cancer, esophageal cancer, gastrointestinal cancer, gastric cancer, colorectal cancer, kidney cancer, clear cell renal carcinoma, head and neck cancer, germ cell cancer, bone cancer, thyroid cancer, skin cancer, neoplasm of the central nervous system, mesothelioma, myeloma, and sarcoma, for example the cancer is colon cancer.

20. A kit comprising a container comprising the polypeptide complex of any of claims 1-11.