

CLAIMS

1. An antibody, wherein

a heavy chain variable region of the antibody comprises HCDR1-HCDR3 with amino acid sequences set forth in SEQ ID NOs: 19–21, respectively, and a light chain variable region of the antibody comprises LCDR1-LCDR3 with amino acid sequences set forth in SEQ ID NOs: 22–24, respectively;

the antibody is of human IgG1 subtype;

wherein, according to the EU numbering system, a heavy chain constant region of the antibody comprises mutations at any 2 or 3 of positions 234, 235 and 237, and an affinity constant of the antibody to FcγRIIIa and/or C1q is reduced after the mutation as compared to that before the mutation; preferably, the affinity constant is measured by a Fortebio Octet system.

2. The antibody according to claim 1, wherein according to the EU numbering system, the heavy chain constant region of the antibody comprises the following mutations:

L234A and L235A;

L234A and G237A;

L235A and G237A;

or

L234A, L235A and G237A.

3. An antibody, wherein

a heavy chain variable region of the antibody comprises HCDR1-HCDR3 with amino acid sequences set forth in SEQ ID NOs: 19–21, respectively, and a light chain variable region of the antibody comprises LCDR1-LCDR3 with amino acid sequences set forth in SEQ ID NOs: 22–24, respectively;

the antibody is of human IgG1 subtype;

wherein according to the EU numbering system, a heavy chain constant region of the antibody comprises the following mutations:

L234A and L235A;

L234A and G237A;

L235A and G237A;

or

L234A, L235A and G237A.

4. The antibody according to any of claims 1–3, wherein:

(a) according to the EU numbering system, the heavy chain constant region of the antibody further comprises one or more mutations selected from:

N297A, D265A, D270A, P238D, L328E, E233D, H268D, P271G, A330R, C226S, C229S, E233P, P331S, S267E, L328F, A330L, M252Y, S254T, T256E, N297Q, P238S, P238A, A327Q, A327G, P329A, K322A, T394D, G236R, G236A, L328R, A330S, P331S, H268A, E318A and K320A; or

~~**5. The antibody according to any of claims 1–4, wherein**~~

(b) the heavy chain variable region of the antibody comprises an amino acid sequence selected from SEQ ID NO: 2 and SEQ ID NO: 6; and the light chain variable region of the antibody comprises an amino acid sequence selected from SEQ ID NO: 4 and SEQ ID NO: 8; or

~~**6. The antibody according to any of claims 1–4, wherein**~~

(c) the heavy chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 2, and the light chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 4;

the heavy chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 2, and the light chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 8;

the heavy chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 6, and the light chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 4;

or

the heavy chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 6, and the light chain variable region of the antibody comprises an amino acid sequence set forth in SEQ ID NO: 8; or

~~7. The antibody according to any of claims 1–6, wherein~~

(d) the heavy chain is set forth in SEQ ID NO: 16, and the light chain is set forth in SEQ ID NO: 12;

or

the heavy chain is set forth in SEQ ID NO: 18, and the light chain is set forth in SEQ ID NO: 12.

~~85.~~ The antibody according to any of claims 1–~~74~~, wherein:

(a) the antibody binds to FcγRIIIa_F158, FcγRI, FcγRIIa_H131, FcγRIIIa_V158 and/or FcγRIIb with an affinity constant greater than about 10^{-7} M, for example, greater than about 10^{-6} M, 10^{-5} M, 10^{-4} M, or 10^{-3} M or greater; preferably, the affinity constant is measured by a Fortebio Octet system;

Preferably, the antibody has no binding signal or a binding signal of less than 0.1 nm to FcγRIIIa_F158, FcγRI, FcγRIIa_H131, FcγRIIIa_V158 and/or FcγRIIb; preferably, the binding signal refers to a response measured by a Fortebio Octet system; or

~~9. The antibody according to any of claims 1–8, wherein (b)~~ the antibody binds to C1q with an affinity constant greater than about 10^{-9} M, for example, greater than about 10^{-8} M, 10^{-7} M, 10^{-6} M, or 10^{-5} M or greater; preferably, the affinity constant is measured by a Fortebio Octet system;

preferably, the antibody has no binding signal or a binding signal of less than 0.1 nm to C1q; preferably, the binding signal refers to a response measured by a Fortebio Octet system.

~~106.~~ An isolated nucleic acid molecule, encoding the antibody according to any of claims 1–~~95~~.

~~117.~~ A vector, comprising the isolated nucleic acid molecule according to claim ~~106~~.

~~128~~. A host cell, comprising the isolated nucleic acid molecule according to claim **~~106~~** or the vector according to claim **~~117~~**.

~~139~~. A conjugate, comprising the antibody according to any of claims 1–**~~95~~** and a conjugated moiety, wherein the conjugated moiety is a detectable label; preferably, the conjugated moiety is a radioisotope, a fluorescent substance, a luminescent substance, a colored substance, or an enzyme.

~~1410~~. A kit, comprising the antibody according to any of claims 1–**~~95~~** or the conjugate according to claim **~~139~~**;

wherein preferably, the kit further comprises a second antibody specifically recognizing the antibody; optionally, the second antibody further comprises a detectable label, for example, a radioisotope, a fluorescent substance, a luminescent substance, a colored substance, or an enzyme.

~~1511~~. Use of the antibody according to any of claims 1–**~~95~~** or the conjugate according to claim **~~139~~** in preparing a kit for detecting the presence or level of PD-1 in a sample.

~~1612~~. A pharmaceutical composition, comprising the antibody according to any of claims 1–**~~95~~** or the conjugate according to claim **~~139~~**, wherein, optionally, the pharmaceutical composition further comprises a pharmaceutically acceptable carrier and/or excipient.

~~1713~~. The pharmaceutical composition according to claim **~~1612~~**, further comprising one or more anti-tumor chemotherapeutics;
preferably, the anti-tumor chemotherapeutic is a tyrosine kinase inhibitor; more preferably, the anti-tumor chemotherapeutic is anlotinib or a pharmaceutically acceptable salt thereof (e.g., hydrochloride salt), or lenvatinib or a pharmaceutically acceptable salt thereof (e.g., mesylate salt); **~~or~~**

~~18. The pharmaceutical composition according to claim 16 or 17,~~ wherein the unit dose of the pharmaceutical composition is 100–1000 mg, 200–800 mg, 200–500 mg, 300–600 mg, 400–500 mg, or 450 mg, based on the mass of the antibody.

~~19~~14. A therapeutic combination, comprising the antibody according to any of claims 1–~~95~~, and at least one (e.g., 1, 2 or 3) anti-tumor chemotherapeutic.

~~20~~15. The therapeutic combination according to claim ~~19~~14, wherein the anti-tumor chemotherapeutic is a tyrosine kinase inhibitor; preferably, the anti-tumor chemotherapeutic is anlotinib or a pharmaceutically acceptable salt thereof (e.g., hydrochloride salt), or lenvatinib or a pharmaceutically acceptable salt thereof (e.g., mesylate salt); ~~or~~

~~21. The therapeutic combination according to claim 19 or 20,~~ wherein the unit dose of the antibody is 100–1000 mg, 200–800 mg, 200–500 mg, 300–600 mg, 400–500 mg, or 450 mg; ~~or~~

~~22. The therapeutic combination according to claim 19 or 20,~~ wherein the unit dose of the anti-tumor chemotherapeutic is 0.1–100 mg, 0.5–50 mg, 1–20 mg, 2–15 mg, 4–12 mg, or 8–12 mg; ~~or~~

~~23. The therapeutic combination according to any of claims 19–22,~~ wherein the therapeutic combination is a fixed combination, e.g., in the form of a solid pharmaceutical composition or a liquid pharmaceutical composition; or the therapeutic combination is a non-fixed combination, e.g., the anti-PD-1 antibody and the anti-tumor chemotherapeutic in the non-fixed combination are each in the form of a pharmaceutical composition.

~~24~~16. A kit product, comprising: the pharmaceutical composition according to ~~any of claims 16–18~~12 or 13 or the therapeutic combination according to ~~any of claims 19–23~~14 or 15, and

a package insert.

2517. Use of the antibody according to any of claims 1–**95**, the conjugate according to claim **139**, the pharmaceutical composition according to ~~any of claims 16–18~~**12 or 13** or the therapeutic combination according to ~~any of claims 19–23~~**14 or 15** in preparing a medicament for treating and/or preventing a tumor or anemia, or in preparing a medicament for diagnosing a tumor or anemia, wherein preferably the tumor is selected from one or more of melanoma, renal cancer, prostate cancer, bladder cancer, colon cancer, rectal cancer, gastric cancer, liver cancer, lung cancer, ovarian cancer, leukemia, nasopharyngeal cancer and endometrial cancer;

preferably, the lung cancer is selected from one or more of non-small cell lung cancer, small cell lung cancer and squamous cell lung cancer;

preferably, the gastric cancer is gastric adenocarcinoma or gastroesophageal junction adenocarcinoma;

preferably, the tumor is a solid tumor of MSI-H/dMMR phenotype; preferably, the tumor is selected from one or more of the following tumors of MSI-H/dMMR phenotype:

colon cancer, rectal cancer, endometrial cancer, gastric cancer, mesothelioma, sarcoma, adrenocortical carcinoma, malignant melanoma and ovarian germ cell neoplasm.

2618. Use of the antibody according to any of claims 1–**95**, the conjugate according to claim **139**, the pharmaceutical composition according to ~~any of claims 16–18~~**12 or 13** or the therapeutic combination according to ~~any of claims 19–23~~**14 or 15** in preparing:

a medicament for blocking the binding of PD-1 to PD-L1,

a medicament for down-regulating the activity or level of PD-1,

a medicament for relieving the immunosuppression of PD-1 in an organism, or

a medicament for elevating IFN- γ and/or IL-2 expression in T lymphocytes.

2719. The antibody according to any of claims 1–**95**, the conjugate according to claim **139**, the pharmaceutical composition according to ~~any of claims 16–18~~**12 or 13** or the therapeutic combination according to ~~any of claims 19–23~~**14 or 15** for use in treating and/or preventing

a tumor or anemia, or for use in diagnosing a tumor or anemia, wherein preferably the tumor is selected from one or more of melanoma, renal cancer, prostate cancer, bladder cancer, colon cancer, rectal cancer, gastric cancer, liver cancer, lung cancer, ovarian cancer, leukemia, nasopharyngeal cancer and endometrial cancer;

preferably, the lung cancer is selected from one or more of non-small cell lung cancer, small cell lung cancer and squamous cell lung cancer;

preferably, the gastric cancer is gastric adenocarcinoma or gastroesophageal junction adenocarcinoma;

preferably, the tumor is a solid tumor of MSI-H/dMMR phenotype; preferably, the tumor is selected from one or more of the following tumors of MSI-H/dMMR phenotype:

colon cancer, rectal cancer, endometrial cancer, gastric cancer, mesothelioma, sarcoma, adrenocortical carcinoma, malignant melanoma and ovarian germ cell neoplasm.

~~28. A method for treating and/or preventing a tumor or anemia, or a method of diagnosing a tumor or anemia, comprising: administering to a subject in need an effective amount of the antibody according to any of claims 1-9, the conjugate according to claim 13, the pharmaceutical composition according to any of claims 16-18 or the therapeutic combination according to any of claims 19-23, wherein preferably the tumor is selected from one or more of melanoma, renal cancer, prostate cancer, bladder cancer, colon cancer, rectal cancer, gastric cancer, liver cancer, lung cancer, ovarian cancer, leukemia, nasopharyngeal cancer and endometrial cancer;~~

~~preferably, the lung cancer is selected from one or more of non-small cell lung cancer, small cell lung cancer and squamous cell lung cancer;~~

~~preferably, the gastric cancer is gastric adenocarcinoma or gastroesophageal junction adenocarcinoma;~~

~~preferably, the tumor is a solid tumor of MSI-H/dMMR phenotype; preferably, the tumor is selected from one or more of the following tumors of MSI-H/dMMR phenotype:~~

~~colon cancer, rectal cancer, endometrial cancer, gastric cancer, mesothelioma, sarcoma, adrenocortical carcinoma, malignant melanoma and ovarian germ cell neoplasm.~~

~~29~~20. The use of claim 17 or 18 or the antibody, the conjugate, the pharmaceutical composition or the therapeutic combination for use of claim 19~~method according to claim 28~~, wherein ~~the~~an effective amount of the antibody is to be administered to ~~the~~a subject in need before or after surgical treatment and/or before or after radiotherapy;; or

~~30. The method according to claim 28 or 29~~, wherein the unit dose of the antibody is 0.1–100 mg per kg body weight, preferably 1–10 mg per kg body weight; alternatively, the unit dose of the antibody is 10–1000 mg, preferably 50–500 mg in each subject; preferably, the dose is to be given once every 3 days, 4 days, 5 days, 6 days, 10 days, 1 week, 2 weeks or 3 weeks; preferably, the route of administration is intravenous drip infusion or intravenous injection;; or

~~31. The method according to any of claims 28–30~~, wherein the administration of the antibody is to be performed in cycles of 2 or 3 weeks, and preferably, the antibody is to be administered intravenously on the first day of each cycle; preferably, the antibody is to be administered once every two or three weeks.