

X₃₄ is G or E or F or H or M or Q or S,

X₃₅ is W or A or I or P or Q or T or V,

X₃₆ is Y or G or N or Q,

X₃₇ is G or S or T, and

X₃₈ is Y or I.

[0008a] In one aspect, the present disclosure refers to a conditionally active anti-Ror2 antibody or antibody fragment that specifically binds to Ror2 protein with a ratio of binding affinity to the Ror protein at a value of a pH of pH 5.8 to pH 6.8 in a tumor microenvironment in comparison with a different value of pH 7.0 to pH 7.6 that occurs in a non-tumor microenvironment of at least 1.5:1, said antibody or antibody fragment comprising a heavy chain variable region and a light chain variable region, said heavy chain variable region comprising three complementarity determining regions having H1, H2, and H3 sequences, wherein:

(a) the H1 sequence is GYTX₁TEX₂X₃X₄H (SEQ ID NO:1);

(b) the H2 sequence is X₅X₆X₇X₈NNGGTGYNQKFKG (SEQ ID NO:2); and

(c) the H3 sequence is X₉X₁₀X₁₁SX₁₂YX₁₃YX₁₄X₁₅SYFX₁₆X₁₇X₁₈ (SEQ ID NO:3);

wherein

X₁ is F or E,

X₂ is Y or D,

X₃ is T or C,

X₄ is M or D or E or Y,

X₅ is G or S,

X₆ is I or E,

X₇ is N or C or L or V,

X₈ is T or D or E,

X₉ is A or I or M or S or T,

X₁₀ is R or Q or H,

X₁₁ is G or E,

X₁₂ is L,

X₁₃ is S,

X₁₄ is G,

X₁₅ is N or E,

X₁₆ is D or L,

X₁₇ is Y or T, and

X₁₈ is W or L, and

said light chain variable region comprising three light chain complementarity determining regions having L1, L2, and L3 sequences, wherein:

- (a) the L1 sequence is SATSSX₁₉X₂₀X₂₁MX₂₂ (SEQ ID NO:7);
- (b) the L2 sequence is X₂₃TSNLAS (SEQ ID NO:8); and
- (c) the L3 sequence is QX₂₄X₂₅SX₂₆YPFX₂₇X₂₈ (SEQ ID NO:9);

wherein

X₁₉ is V or E,

X₂₀ is S or D,

X₂₁ is Y or C or D,

X₂₂ is H or G or L,

X₂₃ is G or C or H or P,

X₂₄ is Q or E,

X₂₅ is R or H,

X₂₆ is S or G or I or Q,

X₂₇ is T or D,

X₂₈ is F or D or E, and

wherein the antibody or antibody fragment has only one single point mutation in one of the heavy and light chain complementarity determining regions H1, H2, H3, L1, L2, and L3, said one single point mutation being determined in comparison with the parent antibody in which X₁ to X₂₈ are F, Y, T, M, G, I, N, T, A, R, G, L, S, G, N, D, Y, W, V, S, Y, H, G, Q, R, S, T, and F, respectively.

[0009] In one example, this isolated heavy chain variable region polypeptide is combined with an isolated light chain variable region that includes three complementarity determining regions L1, L2, and L3 sequences, wherein:

the L1 sequence is SATSSX₁₉X₂₀X₂₁MX₂₂ (SEQ ID NO:7) or RASESVDRYGNSX₃₉IH (SEQ ID NO:10);

the L2 sequence is X₂₃TSNLAS (SEQ ID NO:8) or X₄₀TYX₄₁LES (SEQ ID NO:11); and

the L3 sequence is QX₂₄X₂₅SX₂₆YPFX₂₇X₂₈ (SEQ ID NO:9) or QQX₄₂NX₄₃DPX₄₄TX₄₅ (SEQ ID NO:12);

wherein

X₁₉ is V or E,

X₂₀ is S or D,

X₂₁ is Y or C or D,

X₂₂ is H or G or L,

X₂₃ is G or C or H or P,

X₂₄ is Q or E,

X₂₅ is R or H,

X₂₆ is S or D or G or I or Q or V,

X₂₇ is T or D,

X₂₈ is F or D or E,

X₃₉ is F or S or T,

X₄₀ is R or C or D or E or W,

X₄₁ is N or D,

X₄₂ is T or I or P,

X₄₃ is E or V,

X₄₄ is W or T, and

WHAT IS CLAIMED IS:

1. A conditionally active anti-Ror2 antibody or antibody fragment that specifically binds to Ror2 protein with a ratio of binding affinity to the Ror protein at a value of a pH of pH 5.8 to pH 6.8 in a tumor microenvironment in comparison with a different value of pH 7.0 to pH 7.6 that occurs in a non-tumor microenvironment of at least 1.5:1, said antibody or antibody fragment comprising a heavy chain variable region and a light chain variable region, said heavy chain variable region comprising three complementarity determining regions having H1, H2, and H3 sequences, wherein:

(a) the H1 sequence is GYTX₁TEX₂X₃X₄H (SEQ ID NO:1);

(b) the H2 sequence is X₅X₆X₇X₈NNGGTGYNQKFKG (SEQ ID NO:2); and

(c) the H3 sequence is X₉X₁₀X₁₁SX₁₂YX₁₃YX₁₄X₁₅SYFX₁₆X₁₇X₁₈ (SEQ ID NO:3);

wherein

X₁ is F or E,

X₂ is Y or D,

X₃ is T or C,

X₄ is M or D or E or Y,

X₅ is G or S,

X₆ is I or E,

X₇ is N or C or L or V,

X₈ is T or D or E,

X₉ is A or I or M or S or T,

X₁₀ is R or Q or H,

X₁₁ is G or E,

X₁₂ is L,

X₁₃ is S,

X₁₄ is G,

X₁₅ is N or E,

X₁₆ is D or L,

X₁₇ is Y or T, and

X₁₈ is W or L, and

said light chain variable region comprising three light chain complementarity determining regions having L1, L2, and L3 sequences, wherein:

(a) the L1 sequence is SATSSX₁₉X₂₀X₂₁MX₂₂ (SEQ ID NO:7);

(b) the L2 sequence is X₂₃TSNLAS (SEQ ID NO:8); and

(c) the L3 sequence is QX₂₄X₂₅SX₂₆YPFX₂₇X₂₈ (SEQ ID NO:9);

wherein

X₁₉ is V or E,

X₂₀ is S or D,

X₂₁ is Y or C or D,

X₂₂ is H or G or L,

X₂₃ is G or C or H or P,

X₂₄ is Q or E,

X₂₅ is R or H,

X₂₆ is S or G or I or Q,

X₂₇ is T or D,

X₂₈ is F or D or E, and

wherein the antibody or antibody fragment has only one single point mutation in one of the heavy and light chain complementarity determining regions H1, H2, H3, L1, L2, and L3, said one single point mutation being determined in comparison with the parent antibody in which X₁ to X₂₈ are F, Y, T, M, G, I, N, T, A, R, G, L, S, G, N, D, Y, W, V, S, Y, H, G, Q, R, S, T, and F, respectively.

2. The antibody or antibody fragment of claim 1, wherein the heavy chain variable region has an amino acid sequence selected from sequences of SEQ ID NOS: 18 and 20-22.
3. The antibody or antibody fragment of any one of claims 1-2, wherein the light chain variable region has an amino acid sequence selected from SEQ ID NOS: 13, 15, and 16.
4. The antibody or antibody fragment of claim 1, wherein X₁ to X₂₈ are F, Y, T, M, G, I, N, T, A, H, G, L, S, G, N, D, Y, W, V, S, Y, H, G, Q, R, S, T and F, respectively.
5. The antibody or antibody fragment of claim 1, wherein the heavy chain variable region has an amino acid sequence selected from SEQ ID NOS: 20-22 and the light chain variable region has an amino acid sequence selected from SEQ ID NOS: 15 and 16.
6. The antibody or antibody fragment of claim 5, wherein the heavy chain variable region has the amino acid sequence of SEQ ID NO: 20 and the light chain variable region has the amino acid sequence of SEQ ID NO: 15.

7. The antibody or antibody fragment of claim 5, wherein the heavy chain variable region has the amino acid sequence of SEQ ID NO: 21 and the light chain variable region has the amino acid sequence of SEQ ID NO: 16.
8. The antibody or antibody fragment of claim 5, wherein the heavy chain variable region has the amino acid sequence of SEQ ID NO: 22 and the light chain variable region has the amino acid sequence of SEQ ID NO: 16.
9. The antibody or antibody fragment of any one of claims 1-8, wherein the antibody or antibody fragment has a ratio of binding affinity to the Ror2 protein in the tumor microenvironment to a binding affinity to the Ror2 protein in the non-tumor microenvironment of at least 2:1, at least 3:1, at least 4:1, at least 5:1, at least 6:1, at least 7:1, at least 8:1, at least 9:1, at least 10:1, at least 20:1, at least 30:1, at least 50:1, at least 70:1, or at least 100:1.
10. The antibody or antibody fragment of any one of claims 1-9, wherein the antibody or antibody fragment is a chimeric antibody, a multispecific antibody, or a humanized antibody.
11. An immunoconjugate comprising the antibody or antibody fragment of any one of claims 1-10.
12. The immunoconjugate of claim 11, wherein the immunoconjugate comprises at least one agent selected from a chemotherapeutic agent, a radioactive atom, a cytostatic agent and a cytotoxic agent.
13. The immunoconjugate of claim 12, comprising at least two said agents.
14. The immunoconjugate of any one of claims 12-13, wherein the antibody or antibody fragment and the at least one or two agents are covalently bonded to a linker molecule.

15. The immunoconjugate of any one of claims 12-14, wherein the at least one or two agents are selected from maytansinoids, auristatins, dolastatins, calicheamicin, pyrrolobenzodiazepines, and anthracyclines.
16. A pharmaceutical composition comprising:
the antibody or antibody fragment of any one of claims 1-10, or the immunoconjugate of any one of claims 11-15; and
a pharmaceutically acceptable carrier.
17. The pharmaceutical composition of claim 16, further comprising a tonicity agent.
18. A kit for diagnosis or treatment, said kit comprising the antibody or antibody fragment of any one of claims 1-10, or the immunoconjugate of any one of claims 12-15, or the pharmaceutical composition of any one of claims 16-17 and instructions for using the antibody or antibody fragment, the immunoconjugate and/or the pharmaceutical composition for diagnosis or treatment.