

Claims

1. An isolated antibody or antigen-binding fragment thereof that binds human cannabinoid type 1 receptor (CB1) (e.g. SEQ ID NO:1), the antibody or antigen-binding fragment thereof
5 comprising six complementarity determining regions (CDRs): CDR-H1, CDR-H2, CDR-H3, CDR-L1, CDR-L2, and CDR-L3, wherein

CDR-H1 has an amino acid sequence G-Y-T-F-T-D-Y-W (residues 26-33 of SEQ ID NO:329) or a modification of said amino acid sequence by a substitution of at least one amino acid residue, wherein the substitution of G at position 1 is S; the substitution of T at
10 position 3 is E; the substitution of T at position 5 is S or N; the substitution of D at position 6 is R or Y; the substitution of Y at position 7 is H; and the substitution of W at position 8 is A or N;

CDR-H2 has an amino acid sequence I-Y-P-Y-D-G-D-T (residues 51-58 of SEQ ID NO:329) or a modification of said amino acid sequence by a substitution of at least one
15 amino acid residue, wherein the substitution of I at position 1 is F; the substitution of Y at position 2 is D, S, or T; the substitution of P at position 3 is T; the substitution of Y at position 4 is G, D, or S; the substitution of D at position 5 is Y or S; the substitution of G at position 6 is S; the substitution of D at position 7 is E, G, or R; and the substitution of T at position 8 is A, S, or I;

CDR-H3 has an amino acid sequence A-R-G-X₁-X₂-X₃-X₄-X₅-X₆-X₇-X₈-X₉-W-X₁₀-X₁₁-Y (residues 98-113 of SEQ ID NO:329) or a modification of said amino acid sequence by substitution of at least one amino acid residue, wherein the substitution of A at position 1 is S; the substitution of G at position 3 is S; X₁ at position 4 is Q, Y, K, R, or G, or is not present; X₂ at position 5 is E, Y, L or G, or is not present; X₃ at position 6 is Y or P, or is not
25 present; X₄ at position 7 is Y, R, or E, or is not present; X₅ at position 8 is G, or is not present; X₆ at position 9 is T, or is not present; X₇ at position 10 is N or D, or is not present; X₈ at position 11 is Y, N, A, or G, or is not present; X₉ at position 12 is N, Y, S, A, or R, or is not present; the substitution of W at position 13 is Y, A, or P; X₁₀ at position 14 is L, M, F, or G, or is not present; X₁₁ at position 15 is P, D, A, or T, or is not present; the substitution of Y at position 16 is V;
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CDR-L1 has an amino acid sequence Q-X₁-I-S-S-X₂-Y (residues 27-33 of SEQ ID NO:330) or a modification of said amino acid sequence by substitution of at least one amino acid residue, wherein the substitution of Q at position 1 is S or E; X₁ at position 2 is E, S, T, N, G, or R; the substitution of I at position 3 is V; the substitution of S at position 4 is A, R,

or G; the substitution of S at position 5 is G, N, or T; X₂ at position 6 is S, N, the peptide F-R-Y-S, or is not present; and the substitution of Y at position 7 is F, D, or N;

CDR-L2 has an amino acid sequence: X₁-T-S (residues 51-53 of SEQ ID NO:330) or a modification of said amino acid sequence by substitution of at least one amino acid residue, wherein X₁ at position 1 is A, Y, G, R, D, or S; the substitution of T at position 2 is A; the substitution of S at position 3 is R; and

CDR-L3 has an amino acid sequence: Q-Q-Y-X₁-S-X₂-P-Y-T(residues 91-99 of SEQ ID NO:330) or a modification of said amino acid sequence by substitution of at least one amino acid residue, wherein the substitution of Q at position 1 is L or H; the substitution of Q at position 2 is H; the substitution of Y at position 3 is S or G; X₁ at position 4 is S, W, H, Y, N, or I; the substitution of S at position 5 is E, R, G, T, or N; X₂ at position 6 is Y, I, S, T, L, or W; and the substitution of Y at position 8 is P, L, F, or is not present;

wherein said substitution of at least one amino acid residue does not inhibit the ability of said antibody or antigen-binding fragment thereof to bind human CB1.

~~2. An isolated antibody or antigen-binding fragment thereof that binds human cannabinoid type 1 receptor (CB1) (SEQ ID NO:1) wherein the antibody comprises CDRs of a variable heavy (VH) domain sequence and CDRs of a variable light (VL) domain sequence, wherein the VH domain sequence is selected from the group consisting of SEQ ID NOs: 18, 30, 42, 54, 66, 78, 90, 102, 114, 126, 138, 150, 162, 174, 186, 198, 210, 222, 234, 246, 258, 270, 282, 294, 306, and 318, and wherein the VL domain is selected from the group consisting of SEQ ID NOs: 24, 36, 48, 60, 72, 84, 96, 108, 120, 132, 144, 156, 168, 180, 192, 204, 216, 228, 240, 252, 264, 276, 288, 300, 312, and 324.~~

~~3. An isolated antibody or antigen-binding fragment thereof that binds human cannabinoid type 1 receptor (CB1) (SEQ ID NO:1) wherein the antibody comprises a variable heavy (VH) domain sequence and a variable light (VL) domain sequence, wherein the VH domain sequence is selected from the group consisting of SEQ ID NOs: 18, 30, 42, 54, 66, 78, 90, 102, 114, 126, 138, 150, 162, 174, 186, 198, 210, 222, 234, 246, 258, 270, 282, 294, 306, and 318, and/or wherein the VL domain is selected from the group consisting of SEQ ID NOs: 24, 36, 48, 60, 72, 84, 96, 108, 120, 132, 144, 156, 168, 180, 192, 204, 216, 228, 240, 252, 264, 276, 288, 300, 312, and 324.~~

~~4. The isolated antibody or antigen-binding fragment thereof of claim 3, wherein the~~

antibody or antigen-binding fragment thereof comprises the heavy and light chain CDRs of a VH/VL pair selected from the group consisting of SEQ ID NOs: 18/24, 30/36, 42/48, 54/60, 66/72, 78/84, 90/96, 102/108, 114/120, 126/132, 138/144, 150/156, 162/168, 174/180, 186/192, 198/204, 210/216, 222/228, 234/240, 246/252, 258/264, 270/276, 282/288, 294/300, 306/312, and 318/324.

5. The isolated antibody or antigen-binding fragment thereof of claim 4, wherein the antibody or antigen-binding fragment thereof comprises the VH/VL pair selected from the group consisting of: SEQ ID NOs: 18/24, 30/36, 42/48, 54/60, 66/72, 78/84, 90/96, 102/108, 114/120, 126/132, 138/144, 150/156, 162/168, 174/180, 186/192, 198/204, 210/216, 222/228, 234/240, 246/252, 258/264, 270/276, 282/288, 294/300, 306/312, and 318/324.

6. The isolated antibody or antigen-binding fragment thereof of claim 3, wherein the antibody or antigen-binding fragment thereof comprises an HCDR set (HCDR1, HCDR2, HCDR3) selected from the group consisting of (20, 21, 22); (32, 33, 34); (44, 45, 46); (56, 57, 58); (68, 69, 70); (80, 81, 82); (92, 93, 94); (104, 105, 106); (116, 117, 118); (128, 129, 130); (140, 141, 142); (152, 153, 154); (164, 165, 166); (176, 177, 178); (188, 189, 190); (200, 201, 202); (212, 213, 214); (224, 225, 226); (236, 237, 238); (248, 249, 250); (260, 261, 262); (272, 273, 274); (284, 285, 286); (296, 297, 298); (308, 309, 310); and (320, 321, 322) and an LCDR set (LCDR1, LCDR2, LCDR3) selected from the group consisting of (26, 27, 28); (38, 39, 40); (50, 51, 52); (62, 63, 64); (74, 75, 76); (86, 87, 88); (98, 99, 100); (110, 111, 112); (122, 123, 124); (134, 135, 136); (146, 147, 148); (158, 159, 160); (170, 171, 172); (182, 183, 184); (194, 195, 196); (206, 207, 208); (218, 219, 220); (230, 231, 232); (242, 243, 244); (254, 255, 256); (266, 267, 268); (278, 279, 280); (290, 291, 292); (302, 303, 304); (314, 315, 316); and (326, 327, 328).

7. The isolated antibody or antigen-binding fragment thereof of claim 6, wherein the antibody or antigen-binding fragment thereof comprises the (HCDR set/LCDR set) pair selected from the group consisting of SEQ ID NOs: (20, 21, 22/ 26, 27, 28); (32, 33, 34/ 38, 39, 40); (44, 45, 46/ 50, 51, 52); (56, 57, 58/ 62, 63, 64); (68, 69, 70/ 74, 75, 76); (80, 81, 82/ 86, 87, 88); (92, 93, 94/ 98, 99, 100); (104, 105, 106/ 110, 111, 112); (116, 117, 118/ 122, 123, 124); (128, 129, 130/ 134, 135, 136); (140, 141, 142/ 146, 147, 148); (152, 153, 154/ 158, 159, 160); (164, 165, 166/ 170, 171, 172); (176, 177, 178/ 182, 183, 184); (188, 189, 190/ 194, 195, 196); (200, 201, 202/ 206, 207, 208); (212, 213, 214/ 218, 219, 220); (224,

225, 226/ 230, 231, 232); (236, 237, 238/ 242, 243, 244); (248, 249, 250/ 254, 255, 256); (260, 261, 262/ 266, 267, 268); (272, 273, 274/ 278, 279, 280); (284, 285, 286/ 290, 291, 292); (296, 297, 298/ 302, 303, 304); (308, 309, 310/ 314, 315, 316); and (320, 321, 322/ 326, 327, 328).

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83. The isolated antibody or antigen-binding fragment thereof of claim 71, wherein the antibody or antigen-binding fragment thereof comprises the HC/LC pair selected from the group consisting of SEQ ID NOs: 17/23, 29/35, 41/47, 53/59, 65/71, 77/83, 89/95, 101/107, 113/119, 125/131, 137/143, 149/155, 161/167, 173/179, 185/191, 197/203, 209/215, 221/227, 233/239, 245/251, 257/263, 269/275, 281/287, 283/289, 305/311, and 317/323.

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~~9. An isolated antibody or antigen-binding fragment thereof that binds human cannabinoid type 1 receptor (CB1) (SEQ ID NO:1) wherein the antibody comprises CDRs of a variable heavy (VH) domain sequence and CDRs of a variable light (VL) domain sequence, wherein the VH domain sequence has at least 95% identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 18, 30, 42, 54, 66, 78, 90, 102, 114, 126, 138, 150, 162, 174, 186, 198, 210, 222, 234, 246, 258, 270, 282, 294, 306, and 318; and the VL domain sequence has at least 95% identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 24, 36, 48, 60, 72, 84, 96, 108, 120, 132, 144, 156, 168, 180, 192, 204, 216, 228, 240, 252, 264, 276, 288, 300, 312, and 324.~~

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~~104. The isolated antibody or antigen-binding fragment thereof of claim 91, wherein the antibody or antigen-binding fragment thereof comprises a variable heavy (VH) domain sequence and a variable light (VL) domain sequence, wherein the VH domain sequence is set forth in an amino acid sequence selected from the group consisting of SEQ ID NOs: 114, 126, 138, 150, 162, 90, 186, 198, 210, 222, 234, 246, 258, 270, 282, 294, 306, and 318, and the VL domain sequence is set forth in an amino acid sequence selected from the group consisting of SEQ ID NOs: 120, 132, 144, 156, 168, 96, 192, 204, 216, 228, 240, 252, 264, 276, 288, 300, 312, and 324.~~

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~~11. The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 126 and the VL is set forth in SEQ ID NO: 132.~~

~~12. The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is~~

set forth in SEQ ID NO: 138 and the VL is set forth in SEQ ID NO: 144.

13. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 150 and the VL is set forth in SEQ ID NO: 156.~~

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14. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 162 and the VL is set forth in SEQ ID NO: 168.~~

15. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 174 and the VL is set forth in SEQ ID NO: 180.~~

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16. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 186 and the VL is set forth in SEQ ID NO: 192.~~

17. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 198 and the VL is set forth in SEQ ID NO: 204.~~

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18. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 210 and the VL is set forth in SEQ ID NO: 216.~~

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19. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 222 and the VL is set forth in SEQ ID NO: 228.~~

20. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 234 and the VL is set forth in SEQ ID NO: 240.~~

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21. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 246 and the VL is set forth in SEQ ID NO: 252.~~

22. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 258 and the VL is set forth in SEQ ID NO: 264.~~

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23. ~~The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 270 and the VL is set forth in SEQ ID NO: 276.~~

~~24. The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 282 and the VL is set forth in SEQ ID NO: 288.~~

~~25. The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 294 and the VL is set forth in SEQ ID NO: 300.~~

~~26. The isolated antibody or antigen-binding fragment thereof of claim 9, wherein the VH is set forth in SEQ ID NO: 306 and the VL is set forth in SEQ ID NO: 312.~~

~~27. The isolated antibody or antigen-binding fragment thereof of claim 1, wherein the VH is set forth in SEQ ID NO: 318 and the VL is set forth in SEQ ID NO: 324.~~

~~28. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-27, wherein the antibody is a human or humanized antibody.~~

~~29~~5. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-~~28~~4, wherein the fragment comprises a Fab fragment, a Fab' fragment, a F(ab)₂ fragment or a scFv fragment.

~~30~~6. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-~~29~~5, wherein the antibody or antigen-binding fragment thereof comprises a human Fc region selected from the group consisting of an IgG1, IgG2, IgG3, IgG4, IgA, IgD, IgE, and IgM Fc.

~~31~~7. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-~~29~~5, wherein the antibody or antigen-binding fragment thereof comprises a modified human Fc region.

~~32~~8. The isolated antibody or antigen-binding fragment thereof of claim ~~31~~7, wherein the ~~antibody or antigen-binding fragment thereof comprises a modified human Fc region~~ comprises a mutation selected from the group consisting of L234A/L235A, S228P, A330S, P331S, E233P/L234V/L235A, A327G/A330S/P331S, L234F/L235E/P331S, and N297Q.

~~33~~9. A multispecific binding protein comprising an antigen-binding fragment of any one of

claims 1-~~32~~8.

~~34. The isolated antibody or antigen-binding fragment of any one of claims 1-33, wherein the antibody or antigen-binding fragment thereof inhibits CB1 signaling activity.~~

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~~35. The isolated antibody or antigen-binding fragment of any one of claims 1-33, wherein the antibody or antigen-binding fragment thereof enhances or activates CB1 signaling activity.~~

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~~36. The isolated antibody or antigen-binding fragment of any one of claims 1-33, wherein the antibody or antigen-binding fragment thereof is an inverse agonist to CB1 signaling activity.~~

~~37~~10. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-~~36~~8, wherein the antibody or antigen-binding fragment thereof is a humanized antibody.

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~~38. The isolated antibody or antigen-binding fragment thereof of any one of claims 1-36, wherein the antibody or antigen-binding fragment thereof is a fully human antibody.~~

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~~39. An isolated antibody or antigen-binding fragment thereof that competes for binding to CB1 with an isolated anti-human CB1 antibody or antigen-binding fragment thereof of any one of claims 1-38.~~

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~~40. The isolated antibody or antigen-binding fragment thereof of claim 39, wherein the antibody or antigen-binding fragment thereof that competes for binding is a human or humanized antibody.~~

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~~41. An antibody or antigen-binding fragment thereof that specifically binds to substantially the same epitope of CB1 as the antibody or antigen-binding fragment of any one of claims 1-38.~~

~~42. The isolated antibody or antigen-binding fragment thereof of claim 41, wherein the antibody or antigen-binding fragment thereof that specifically binds to substantially the same epitope of CB1 is a human or humanized antibody.~~

43. ~~An isolated antibody or antigen-binding fragment thereof that binds to cannabinoid type-1 receptor (CB1), wherein the antibody or antigen-binding fragment thereof has a binding affinity K_d for CB1 of about 1 μ M or less.~~
- 5 44. ~~The isolated antibody or antigen-binding fragment of claim 43, wherein the antibody or antigen-binding fragment thereof has a binding affinity K_d for CB1 of about 100 nM or less.~~
- 10 45. ~~The isolated antibody or antigen-binding fragment of any one of claims 1-44, wherein the antibody or antigen-binding fragment exhibits reduced brain penetration as compared to rimonabant.~~
- 15 46. ~~The isolated antibody or antigen-binding fragment of any one of claims 1-44, wherein the antibody or antigen-binding fragment inhibits CB1 signaling that is at least 2 fold higher as compared to rimonabant.~~
- 20 4811. An isolated nucleic acid molecule encoding an antibody or antigen-binding fragment thereof of any one of claims 1-~~47~~8 and 10 or the multispecific binding protein of claim 9.
4912. An expression vector comprising the nucleic acid molecule of claim 4811.
- 25 5013. An isolated host cell comprising the expression vector of claim 4912.
- 30 51. ~~An *in vitro* method of modulating CB1 signaling, the method comprising contacting a cell expressing CB1 with the antibody or antigen-binding fragment thereof of any one of claims 1-47.~~
52. ~~An *in vitro* method of antagonizing CB1, the method comprising contacting a cell expressing CB1 with the antibody or antigen-binding fragment thereof of any one of claims 1-47.~~

~~53. An *in vitro* method of agonizing CB1, the method comprising contacting a cell expressing CB1 with the antibody or antigen-binding fragment thereof of any one of claims 1-47.~~

5 ~~54. An *in vitro* method of inverse agonizing CB1, the method comprising contacting a cell expressing CB1 with the antibody or antigen-binding fragment thereof of any one claims 1-47.~~

10 ~~55~~14. A pharmaceutical composition comprising an isolated antibody or antigen-binding fragment thereof of any one of claims 1-47~~8~~ and 10 or the multispecific binding protein of claim 9 and at least one pharmaceutically acceptable excipient.

15 ~~56. — The antibody or antigen-binding fragment thereof of any one of claims 1-47, for use in medicine.~~

~~57. — The antibody or antigen-binding fragment thereof of any one of claims 1-47, for use in diagnosing a disease or disorder associated with CB1~~

20 ~~158. Use of the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47~~8 and 10 or the multispecific binding protein of claim 9 in the preparation of a pharmaceutical composition for ~~a method for inhibiting the biological activity of CB1 in a subject in need thereof~~treating a disease or disorder selected from the group consisting of obesity, syndromic obesities including Prader-Willi syndrome (PWS), Alström syndrome, Bardet-Biedel syndrome (BBS), Albright Hereditary Osteodystrophy (AHO), and SIM1

25 deletion syndrome; diabetes and related complications; dyslipidemia; liver diseases such as, for example, non-alcoholic steatohepatitis (NASH), non-alcoholic fatty liver disease, and primary biliary cirrhosis; fibrosis, for example, kidney fibrosis; chronic kidney disease; diabetic neuropathy, focal segmental glomerulosclerosis, renal disease; metabolic diseases, osteoporosis, atherosclerosis, inflammatory disease, cardiovascular disease, cancer, pain,

30 systemic sclerosis, multiple sclerosis spasticity, glaucoma, and nicotine addiction.

~~59. Use of the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47 in the preparation of a pharmaceutical composition for treating a disease associated with CB1 activity.~~

~~60. Use of the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47 in the preparation of a pharmaceutical composition for treating a disease or disorder responsive to modulation of CB1 signaling in a subject in need thereof.~~

~~61. Use of the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47 in the preparation of a pharmaceutical composition for treating a disease or disorder responsive to antagonism or inverse agonism of CB1 signaling in a subject in need thereof.~~

~~62. Use of the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47 in the preparation of a pharmaceutical composition for treating a disease or disorder responsive to agonism of CB1 signaling in a subject in need thereof.~~

~~63. An *in vitro* method for diagnosing a disease or disorder associated with CB1, the method comprising contacting a cell with an antibody or antigen-binding fragment thereof of any one of claims 1-47.~~

~~64. The method of claim 63, wherein the disease or disorder is selected from the group consisting of obesity, syndromic obesities including Prader-Willi syndrome (PWS), Alström syndrome, Bardet-Biedel syndrome (BBS), Albright Hereditary Osteodystrophy (AHO), and SIM1 deletion syndrome; diabetes and related complications; dyslipidemia; liver diseases such as, for example, non-alcoholic steatohepatitis (NASH), non-alcoholic fatty liver disease, and primary biliary cirrhosis; fibrosis, for example, kidney fibrosis; chronic kidney disease; diabetic neuropathy, focal segmental glomerulosclerosis, renal disease; metabolic diseases, osteoporosis, atherosclerosis, inflammatory disease, cardiovascular disease, cancer, pain, systemic sclerosis, multiple sclerosis spasticity, glaucoma, and nicotine addiction.~~

165. An antibody conjugate comprising the isolated antibody or antigen-binding fragment thereof of any one of claims 1-47 and 10 or the multispecific binding protein of claim 9, wherein the antibody or antigen-binding fragment thereof is conjugated to an agent selected from the group consisting of a therapeutic agent, a cytotoxic agent, an immunoadhesion molecule, and an imaging agent.