

WHAT IS CLAIMED:

1. An isolated antibody that specifically binds to human CD96, the antibody comprising a heavy chain variable region (VH) comprising complementarity determining regions (CDRs) CDRH1, CDRH2, and CDRH3, and a light chain variable region (VL) comprising CDRs CDRL1, CDRL2, and CDRL3, wherein:

(a) CDRH1 comprises the amino acid sequence of $X_1YX_2X_3X_4$ (SEQ ID NO: 135), wherein

X_1 is Q or S;

X_2 is A or S;

X_3 is M or I; and

X_4 is H or S;

(b) CDRH2 comprises the amino acid sequence of $X_1IX_2X_3X_4X_5X_6X_7X_8X_9YX_{10}QKFQG$ (SEQ ID NO: 137), wherein

X_1 is W or G;

X_2 is N or I;

X_3 is A, E, V, or P;

X_4 is V, G, W, or I;

X_5 is S, Y, T, N, or F;

X_6 is G or W;

X_7 is D, Y, N, or T;

X_8 is T or A;

X_9 is K or N; and

X_{10} is S or A;

(c) CDRH3 comprises the amino acid sequence of $NWGX_1SYGX_2DV$ (SEQ ID NO: 180), $GYDSRPLDV$ (SEQ ID NO: 19), or $GYDSRPLDY$ (SEQ ID NO: 20), wherein

X_1 is M or L; and

X_2 is M or L;

(d) CDRL1 comprises the amino acid sequence of $RASQSIX_1X_2YLN$ (SEQ ID NO: 139) or $GGNNIGSKIVH$ (SEQ ID NO: 26), wherein

X_1 is S, T, or L; and

X_2 is S, P, or W;

(e) CDRL2 comprises the amino acid sequence of X_1X_2SSLQS (SEQ ID NO: 141) or $DDRDRPS$ (SEQ ID NO: 32), wherein

X_1 is S or A; and

X₂ is A, S, or E; and/or

(f) CDRL3 comprises the amino acid sequence of QQX₁YSTPALX₂ (SEQ ID NO: 143) or QVWDINVHHVI (SEQ ID NO: 35), wherein

X₁ is S or A; and

X₂ is T or S.

2. The isolated antibody of claim 1, wherein:

(a) CDRH1 comprises the amino acid sequence of X₁YX₂MH (SEQ ID NO: 136), wherein

X₁ is Q or S; and

X₂ is A or S;

(b) CDRH2 comprises the amino acid sequence of WINX₁X₂X₃X₄X₅TKYSQKFQG (SEQ ID NO: 138), wherein

X₁ is A, V, or E;

X₂ is V, W, or G;

X₃ is S, Y, T, or N;

X₄ is G or W; and

X₅ is D, N, Y, or T;

(c) CDRH3 comprises the amino acid sequence of NWGX₁SYGX₂DV (SEQ ID NO: 180), wherein

X₁ is M or L; and

X₂ is M or L;

(d) CDRL1 comprises the amino acid sequence of RASQSIX₁X₂YLN (SEQ ID NO: 139), wherein

X₁ is S, T, or L; and

X₂ is S, P, or W;

(e) CDRL2 comprises the amino acid sequence of X₁X₂SSLQS (SEQ ID NO: 141), wherein

X₁ is S or A; and

X₂ is A, S, or E; and/or

(f) CDRL3 comprises the amino acid sequence of QQSYSTPALT (SEQ ID NO: 33) or QQAYSTPALS (SEQ ID NO: 34).

3. The isolated antibody of claim 1, wherein:

(a) CDRH1 comprises the amino acid sequence of SEQ ID NO: 4;

(b) CDRH2 comprises the amino acid sequence of SEQ ID NO: 17;

- (c) CDRH3 comprises the amino acid sequence of SEQ ID NO: 19 or 20;
- (d) CDRL1 comprises the amino acid sequence of SEQ ID NO: 26;
- (e) CDRL2 comprises the amino acid sequence of SEQ ID NO: 32; and/or
- (f) CDRL3 comprises the amino acid sequence of SEQ ID NO: 35.

4. The isolated antibody of claim 1, wherein CDRH1, CDRH2, and CDRH3 comprise the amino acid sequences of SEQ ID NOs: 1, 5, and 18; 2, 6, and 18; 2, 8, and 18; 2, 9, and 18; 2, 10, and 18; 1, 7, and 18; 2, 11, and 18; 1, 12, and 18; 1, 13, and 18; 1, 14, and 18; 3, 15, and 18; 1, 16, and 18; 1, 5, and 140; 1, 5, and 142; 1, 5, and 179; 4, 17, and 19; or 4, 17, and 20, respectively.

5. The isolated antibody of claim 1, wherein CDRL1, CDRL2, and CDRL3 comprise the amino acid sequences of SEQ ID NOs: 21, 28, and 33; 21, 29, and 33; 21, 30, and 33; 21, 31, and 33; 22, 29, and 33; 24, 29, and 33; 23, 29, and 33; 25, 28, and 34; or 26, 32, and 35, respectively.

6. The isolated antibody of claim 1, wherein CDRH1, CDRH2, CDRH3, CDRL1, CDRL2, and CDRL3 comprise the amino acid sequences of SEQ ID NOs: 1, 5, 18, 21, 28, and 33; 1, 5, 18, 21, 29, and 33; 1, 5, 18, 22, 29, and 33; 1, 5, 18, 23, 29, and 33; 1, 5, 18, 24, 29, and 33; 1, 5, 18, 25, 28, and 34; 1, 5, 140, 21, 28, and 33; 1, 5, 142, 21, 28, and 33; 1, 5, 179, 21, 28, and 33; 1, 7, 18, 21, 29, and 33; 1, 12, 18, 21, 28, and 33; 1, 13, 18, 21, 28, and 33; 1, 14, 18, 21, 28, and 33; 1, 16, 18, 21, 28, and 33; 2, 6, 18, 21, 29, and 33; 2, 8, 18, 21, 29, and 33; 2, 9, 18, 21, 30, and 33; 2, 10, 18, 21, 29, and 33; 2, 11, 18, 21, 31, and 33; 3, 15, 18, 21, 28, and 33; 4, 17, 19, 26, 32, and 35; or 4, 17, 20, 26, 32, and 35, respectively.

7. The isolated antibody of any one of claims 1-6, wherein the antibody comprises a VH comprising an amino acid sequence that is at least 75%, 80%, 85%, 90%, 95%, 99%, or 100% identical to the amino acid sequence of SEQ ID NO: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61.

8. The isolated antibody of claim 7, wherein the amino acid sequence of the VH consists of the amino acid sequence of SEQ ID NO: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61.

9. The isolated antibody of claim 7 or 8, wherein the X in any one of SEQ ID NOs: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61 is glutamine.
10. The isolated antibody of claim 7 or 8, wherein the X in any one of SEQ ID NOs: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61 is pyroglutamate.
11. The isolated antibody of any one of claims 1-10, wherein the antibody comprises a VL comprising an amino acid sequence that is at least 75%, 80%, 85%, 90%, 95%, 99%, or 100% identical to the amino acid sequence of SEQ ID NO: 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, or 75.
12. The isolated antibody of claim 11, wherein the amino acid sequence of the VL consists of the amino acid sequence of SEQ ID NO: 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, or 75.
13. An isolated antibody that specifically binds to human CD96, the antibody comprising: a VH comprising the amino acid sequence of SEQ ID NO: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61; and/or a VL comprising the amino acid sequence of SEQ ID NO: 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, or 75.
14. The isolated antibody of claim 13, wherein the VH and VL comprise the amino acid sequences of SEQ ID NOs: 36 and 62; 37 and 62; 37 and 63; 37 and 66; 37 and 67; 37 and 68; 37 and 69; 38 and 63; 39 and 63; 40 and 63; 41 and 63; 42 and 63; 43 and 64; 44 and 64; 45 and 63; 46 and 63; 47 and 65; 48 and 62; 49 and 62; 50 and 62; 51 and 62; 52 and 62; 53 and 62; 54 and 62; 55 and 62; 56 and 62; 57 and 62; 58 and 62; 59 and 62; 60 and 70; 60 and 71; 60 and 72; 60 and 73; 60 and 74; 60 and 75; or 61 and 70, respectively.
15. The isolated antibody of claim 14, wherein the amino acid sequences of the VH and VL consist of the amino acid sequences of SEQ ID NOs: 36 and 62; 37 and 62; 37 and 63; 37 and 66; 37 and 67; 37 and 68; 37 and 69; 38 and 63; 39 and 63; 40 and 63; 41 and 63; 42 and 63; 43 and 64; 44 and 64; 45 and 63; 46 and 63; 47 and 65; 48 and 62; 49 and 62; 50 and 62; 51 and 62; 52 and 62; 53 and 62; 54 and 62; 55 and 62; 56 and 62; 57 and 62; 58 and 62; 59 and 62; 60 and 70; 60 and 71; 60 and 72; 60 and 73; 60 and 74; 60 and 75; or 61 and 70, respectively.

16. The isolated antibody of any one of claim 13-15, wherein the X in any one of SEQ ID NOs: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61 is glutamine.
17. The isolated antibody of any one of claim 13-15, wherein the X in any one of SEQ ID NOs: 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, or 61 is pyroglutamate.
18. The isolated antibody of any one of claims 1-17, wherein the antibody specifically binds to the amino acid sequence of SEQ ID NO: 130 or 131.
19. An isolated antibody that specifically binds to the amino acid sequence of SEQ ID NO: 130 or 131.
20. The isolated antibody of claim 18 or 19, wherein the antibody binds to the amino acid sequence of SEQ ID NO: 134.
21. The isolated antibody of any one of claims 1-20, wherein the antibody is internalized upon binding to cells expressing human CD96.
22. An isolated antibody that specifically binds to human CD96, wherein the antibody is internalized upon binding to cells expressing human CD96.
23. The isolated antibody of any one of claims 1-22, wherein the antibody comprises a heavy chain constant region selected from the group consisting of human IgG1, IgG2, IgG3, IgG4, IgA1, and IgA2.
24. The isolated antibody of claim 23, wherein the antibody comprises an IgG1 heavy chain constant region.
25. The isolated antibody of claim 24, wherein the amino acid sequence of the IgG1 heavy chain constant region comprises an N297A mutation, numbered according to the EU numbering system.
26. The isolated antibody of claim 25, wherein the antibody comprises a heavy chain constant region comprising the amino acid sequence of SEQ ID NO: 124 or 176.

27. The isolated antibody of claim 24, wherein the amino acid sequence of the IgG1 heavy chain constant region comprises S239D, A330L, and I332E mutations, numbered according to the EU numbering system.

28. The isolated antibody of claim 27, wherein the antibody comprises a heavy chain constant region comprising the amino acid sequence of SEQ ID NO: 125 or 177.

29. The isolated antibody of claim 24, wherein the amino acid sequence of the IgG1 heavy chain constant region comprises S267E and L328F mutations, numbered according to the EU numbering system.

30. The isolated antibody of claim 29, wherein the antibody comprises a heavy chain constant region comprising the amino acid sequence of SEQ ID NO: 126 or 178.

31. The isolated antibody of any one of claims 1-24, wherein the antibody comprises a heavy chain constant region that is a variant of a wild type heavy chain constant region, wherein the variant heavy chain constant region binds to an Fc γ R with higher affinity than the wild type heavy chain constant region binds to the Fc γ R.

32. The isolated antibody of claim 31, wherein the Fc γ R is Fc γ RIIB.

33. The isolated antibody of any one of claims 1-24, wherein the antibody comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, or 169.

34. The isolated antibody of claim 33, wherein the amino acid sequence of the heavy chain consists of the amino acid sequence of SEQ ID NO: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, or 169.

35. The isolated antibody of claims 33 or 34, wherein the X in any one of SEQ ID NOs: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101,

144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, and 169 is glutamine.

36. The isolated antibody of claims 33 or 34, wherein the X in any one of SEQ ID NOs: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, and 169 is pyroglutamate.

37. The isolated antibody of any one of the preceding claims, wherein the antibody comprises a light chain constant region comprising the amino acid sequence of SEQ ID NO: 122 or 123.

38. The isolated antibody of any one of the preceding claims, wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, or 115.

39. The isolated antibody of claim 38, wherein the amino acid sequence of the light chain consists of the amino acid sequence of SEQ ID NOs: 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, or 115.

40. An isolated antibody that specifically binds to human CD96, the antibody comprising: a heavy chain comprising the amino acid sequence of SEQ ID NO: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, or 169; and/or a light chain comprising the amino acid sequence of SEQ ID NO: 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, or 115.

41. The isolated antibody of claim 40, wherein the amino acid sequence of the heavy chain consists of the amino acid sequence of SEQ ID NO: 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 144, 145, 146, 147, 148, 149, 150, 151, 152, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, or 169; and/or the amino acid sequence of the light chain consists of the amino acid sequence of SEQ ID NO: 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, or 115.

42. The isolated antibody of claim 40, wherein the heavy chain and light chain comprise the amino acid sequences of SEQ ID NOs: 76 and 102; 79 and 103; 78 and 103; 82 and 103; 84 and 104; 83 and 104; 86 and 103; 85 and 103; 81 and 103; 80 and 103; 87 and 105; 77 and 102; 88 and 102; 77 and 106; 77 and 107; 77 and 108; 77 and 103; 89 and 102; 90 and 102; 91 and 102; 92 and 102; 93 and 102; 77 and 109; 94 and 102; 95 and 102; 96 and 102; 97 and 102; 98 and 102; 99 and 102; 100 and 110; 100 and 111; 100 and 112; 100 and 113; 100 and 114; 100 and 115; 101 and 110; 144 and 102; 147 and 103; 146 and 103; 150 and 103; 152 and 104; 151 and 104; 154 and 103; 153 and 103; 149 and 103; 148 and 103; 155 and 105; 145 and 102; 156 and 102; 145 and 106; 145 and 107; 145 and 108; 145 and 103; 157 and 102; 158 and 102; 159 and 102; 160 and 102; 161 and 102; 145 and 109; 162 and 102; 163 and 102; 164 and 102; 165 and 102; 166 and 102; 167 and 102; 168 and 110; 168 and 111; 168 and 112; 168 and 113; 168 and 114; 168 and 115; or 169 and 110, respectively.

43. The isolated antibody of claim 41, wherein the amino acid sequences of the heavy chain and the light chain consist of the amino acid sequences of SEQ ID NOs: 76 and 102; 79 and 103; 78 and 103; 82 and 103; 84 and 104; 83 and 104; 86 and 103; 85 and 103; 81 and 103; 80 and 103; 87 and 105; 77 and 102; 88 and 102; 77 and 106; 77 and 107; 77 and 108; 77 and 103; 89 and 102; 90 and 102; 91 and 102; 92 and 102; 93 and 102; 77 and 109; 94 and 102; 95 and 102; 96 and 102; 97 and 102; 98 and 102; 99 and 102; 100 and 110; 100 and 111; 100 and 112; 100 and 113; 100 and 114; 100 and 115; 101 and 110; 144 and 102; 147 and 103; 146 and 103; 150 and 103; 152 and 104; 151 and 104; 154 and 103; 153 and 103; 149 and 103; 148 and 103; 155 and 105; 145 and 102; 156 and 102; 145 and 106; 145 and 107; 145 and 108; 145 and 103; 157 and 102; 158 and 102; 159 and 102; 160 and 102; 161 and 102; 145 and 109; 162 and 102; 163 and 102; 164 and 102; 165 and 102; 166 and 102; 167 and 102; 168 and 110; 168 and 111; 168 and 112; 168 and 113; 168 and 114; 168 and 115; or 169 and 110, respectively.

44. The isolated antibody of any one of claim 40-43, wherein the X in any one of SEQ ID NOs: 76-101 or 144-169 is glutamine.

45. The isolated antibody of any one of claim 40-43, wherein the X in any one of SEQ ID NOs: 76-101 or 144-169 is pyroglutamate.

46. An isolated antibody that specifically binds to human CD96, wherein the antibody binds to the same epitope of human CD96 as the antibody of any one of the preceding claims.

47. An isolated antibody that specifically binds to human CD96, wherein the antibody competes for binding to human CD96 with the antibody of any one of the preceding claims.
48. The isolated antibody of any one of the preceding claims, wherein the antibody is a human antibody.
49. The isolated antibody of any one of the preceding claims, wherein the antibody is a multispecific antibody.
50. The isolated antibody of any one of the preceding claims, wherein the antibody is conjugated to a cytotoxic agent, cytostatic agent, toxin, radionuclide, or detectable label.
51. The isolated antibody of any one of the preceding claims, wherein the antibody is conjugated to a second antibody.
52. An isolated polynucleotide encoding a VH and/or a VL of the antibody of any one of the preceding claims.
53. A vector comprising the polynucleotide of claim 52.
54. A recombinant host cell comprising the polynucleotide of claim 52 or the vector of claim 55.
55. A pharmaceutical composition comprising the antibody of any one of claims 1-51, the polynucleotide of claim 52, the vector of claim 53, or the host cell of claim 54; and a pharmaceutically acceptable carrier or excipient.
56. A method of producing an antibody that specifically binds to human CD96, the method comprising culturing the host cell of claim 54 under suitable conditions so that the polynucleotide is expressed and the antibody is produced.
57. A method of increasing an immune response in a subject, the method comprising administering to the subject an effective amount of the antibody of any one of claims 1-51, the polynucleotide of claim 52, the vector of claim 53, the host cell of claim 54, or the pharmaceutical composition of claim 55.

58. A method of treating cancer in a subject, the method comprising administering to the subject an effective amount of the antibody of any one of claims 1-51, the polynucleotide of claim 52, the vector of claim 53, the host cell of claim 54, or the pharmaceutical composition of claim 55.
59. The method of claim 57 or 58, wherein the antibody, polynucleotide, vector, host cell, or pharmaceutical composition is administered, systemically, intravenously, subcutaneously, or intratumorally, or is delivered to a tumor draining lymph node.
60. The method of any one of claims 57-59, further comprising administering an additional therapeutic agent to the subject.
61. The method of claim 60, wherein the additional therapeutic agent is a chemotherapeutic agent.
62. The method of claim 61, wherein the additional therapeutic agent is a checkpoint targeting agent.
63. The method of claim 62, wherein the checkpoint targeting agent is selected from the group consisting of an antagonist anti-PD-1 antibody, an antagonist anti-PD-L1 antibody, an antagonist anti-PD-L2 antibody, an antagonist anti-CTLA-4 antibody, an antagonist anti-TIM-3 antibody, an antagonist anti-LAG-3 antibody, an antagonist anti-VISTA antibody, an antagonist anti-TIGIT antibody, an antagonist anti-CEACAM1 antibody, an antagonist anti-CD96 antibody, an agonist anti-GITR antibody, and an agonist anti-OX40 antibody.
64. The method of claim 63, wherein the additional therapeutic agent is an anti-PD-1 antibody, optionally wherein the anti-PD-1 antibody is pembrolizumab or nivolumab.
65. The method of claim 60, wherein the additional therapeutic agent is an inhibitor of indoleamine-2,3-dioxygenase (IDO).
66. The method of claim 65, wherein the inhibitor is selected from the group consisting of epacadostat, F001287, indoximod, and NLG919.
67. The method of claim 60, wherein the additional therapeutic agent is a vaccine.

68. The method of claim 67, wherein the vaccine comprises a heat shock protein peptide complex (HSPPC) comprising a heat shock protein complexed with an antigenic peptide.

69. The method of claim 68, wherein the heat shock protein is hsc70 and is complexed with a tumor-associated antigenic peptide.

70. The method of claim 68, wherein the heat shock protein is gp96 protein and is complexed with a tumor-associated antigenic peptide, optionally wherein the HSPPC is derived from a tumor obtained from a subject.

71. A method of treating an infectious disease in a subject, the method comprising administering to the subject an effective amount of the antibody of any one of claims 1-51, the polynucleotide of claim 52, the vector of claim 53, the host cell of claim 54, or the pharmaceutical composition of claim 55.