

## CLAIMS

1. An anti-CD73 antibody or an antigen-binding portion thereof, wherein the antibody binds to the same epitope of human CD73 as an antibody comprising:
- 5 a) a heavy chain (HC) comprising the amino acid sequences of SEQ ID NOs: 9 and 41 and a light chain (LC) comprising the amino acid sequences of SEQ ID NOs: 13 and 42;
- b) an HC comprising the amino acid sequences of SEQ ID NOs: 10 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs: 14 and 42;
- 10 c) an HC comprising the amino acid sequences of SEQ ID NOs: 11 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs: 15 and 42; or
- d) an HC comprising the amino acid sequences of SEQ ID NOs: 12 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs: 16 and 42.
- 15
2. The anti-CD73 antibody or antigen-binding portion of claim 1, wherein
- a) the heavy chain of said antibody comprises:
- 20 i) heavy chain complementarity determining regions (H-CDR)-1-3 comprising the amino acid sequences of SEQ ID NOs: 17-19, respectively;
- ii) a heavy chain variable domain (VH) comprising an amino acid sequence at least 90% identical to the amino acid sequence of SEQ ID NO: 9;
- 25 iii) a VH comprising the amino acid sequence of SEQ ID NO: 9; or
- iv) a heavy chain (HC) comprising the amino acid sequences of SEQ ID NOs: 9 and 41; and
- b) the light chain of said antibody comprises:
- 30 i) light chain complementarity determining regions (L-CDR)-1-3 comprising the amino acid sequences of SEQ ID NOs: 20-22, respectively;
- ii) a light chain variable domain (VL) comprising an amino acid

- sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 13;
- iii) a VL comprising the amino acid sequence of SEQ ID NO: 13; or
- iv) a light chain (LC) comprising the amino acid sequences of SEQ  
ID NOs: 13 and 42.
- 5
3. The anti-CD73 antibody or antigen-binding portion of claim 1, wherein
- a) the heavy chain of said antibody comprises:
- i) heavy chain complementarity determining regions (H-CDR)-1-3  
comprising the amino acid sequences of SEQ ID NOs: 23-25,  
10 respectively;
- ii) a heavy chain variable domain (VH) comprising an amino acid  
sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 10;
- iii) a VH comprising the amino acid sequence of SEQ ID NO: 10; or
- 15 iv) a heavy chain (HC) comprising the amino acid sequences of  
SEQ ID NOs: 10 and 41; and
- b) the light chain of said antibody comprises:
- i) light chain complementarity determining regions (L-CDR)-1-3  
comprising the amino acid sequences of SEQ ID NOs: 26-28,  
20 respectively;
- ii) a light chain variable domain (VL) comprising an amino acid  
sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 14;
- iii) a VL comprising the amino acid sequence of SEQ ID NO: 14; or
- 25 iv) a light chain (LC) comprising the amino acid sequences of SEQ  
ID NOs: 14 and 42.
4. The anti-CD73 antibody or antigen-binding portion of claim 1, wherein
- a) the heavy chain of said antibody comprises:
- i) heavy chain complementarity determining regions (H-CDR)-1-3  
30 comprising the amino acid sequences of SEQ ID NOs: 29-31,  
respectively;
- ii) a heavy chain variable domain (VH) comprising an amino acid

- sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 11;
- iii) a VH comprising the amino acid sequence of SEQ ID NO: 11; or
- iv) a heavy chain (HC) comprising the amino acid sequences of  
SEQ ID NOs: 11 and 41; and
- 5 b) the light chain of said antibody comprises:
- i) light chain complementarity determining regions (L-CDR)-1-3  
comprising the amino acid sequences of SEQ ID NOs: 32-34,  
respectively;
- 10 ii) a light chain variable domain (VL) comprising an amino acid  
sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 15;
- iii) a VL comprising the amino acid sequence of SEQ ID NO: 15; or
- iv) a light chain (LC) comprising the amino acid sequences of SEQ  
ID NOs: 15 and 42.
- 15
5. The anti-CD73 antibody or antigen-binding portion of claim 1, wherein
- a) the heavy chain of said antibody comprises:
- i) heavy chain complementarity determining regions (H-CDR)-1-3  
comprising the amino acid sequences of SEQ ID NOs: 35-37,  
respectively;
- 20 ii) a heavy chain variable domain (VH) comprising an amino acid  
sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 12;
- iii) a VH comprising the amino acid sequence of SEQ ID NO: 12; or
- 25 iv) a heavy chain (HC) comprising the amino acid sequences of  
SEQ ID NOs: 12 and 41; and
- b) the light chain of said antibody comprises:
- i) light chain complementarity determining regions (L-CDR)-1-3  
comprising the amino acid sequences of SEQ ID NOs: 38-40,  
respectively;
- 30 ii) a light chain variable domain (VL) comprising an amino acid  
sequence at least 90% identical to the amino acid sequence of  
SEQ ID NO: 16;

- iii) a VL comprising the amino acid sequence of SEQ ID NO: 16; or
  - iv) a light chain (LC) comprising the amino acid sequences of SEQ ID NOs: 16 and 42.
6. An anti-CD73 antibody or an antigen-binding portion thereof, wherein said antibody comprises the H-CDR1-3 and L-CDR1-3 amino acid sequences of:
- a) SEQ ID NOs: 17-22, respectively;
  - b) SEQ ID NOs: 23-28, respectively;
  - c) SEQ ID NOs: 29-34, respectively; or
  - d) SEQ ID NOs: 35-40, respectively.
7. An anti-CD73 antibody or an antigen-binding portion thereof, wherein said antibody comprises a heavy chain variable domain amino acid sequence and a light chain variable domain amino acid sequence that are at least 90% identical to the amino acid sequences of:
- a) SEQ ID NOs: 9 and 13, respectively;
  - b) SEQ ID NOs: 10 and 14, respectively;
  - c) SEQ ID NOs: 11 and 15, respectively; or
  - d) SEQ ID NOs: 12 and 16, respectively.
8. An anti-CD73 antibody or an antigen-binding portion thereof, wherein said antibody comprises a heavy chain variable domain and a light chain variable domain comprising the amino acid sequences of:
- a) SEQ ID NOs: 9 and 13, respectively;
  - b) SEQ ID NOs: 10 and 14, respectively;
  - c) SEQ ID NOs: 11 and 15, respectively; or
  - d) SEQ ID NOs: 12 and 16, respectively.
9. An anti-CD73 antibody that comprises:
- a) a heavy chain (HC) comprising the amino acid sequences of SEQ ID NOs: 9 and 41 and a light chain (LC) comprising the amino acid sequences of SEQ ID NOs: 13 and 42;
  - b) an HC comprising the amino acid sequences of SEQ ID NOs: 10 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs:

- 14 and 42;
- c) an HC comprising the amino acid sequences of SEQ ID NOs: 11 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs: 15 and 42; or
- 5 d) an HC comprising the amino acid sequences of SEQ ID NOs: 12 and 41 and an LC comprising the amino acid sequences of SEQ ID NOs: 16 and 42.
10. An anti-CD73 antibody or an antigen-binding portion thereof that binds to an epitope on human CD73 comprising:
- 10 a) amino acid residues R73, R109, and D168 of SEQ ID NO: 43;
- b) amino acid residue R109 of SEQ ID NO: 43; or
- c) amino acid residues I301, S302, and H304 of SEQ ID NO: 43.
11. An anti-CD73 antibody or an antigen-binding portion thereof that binds to an epitope on human CD73 comprising:
- 15 a) amino acid residues 27-31, 61-75, and 161-170 of SEQ ID NO: 43;
- b) amino acid residues 61-70 and 161-170 of SEQ ID NO: 43; or
- c) amino acid residues 27-31, 266-270, and 291-305 of SEQ ID NO: 43.
12. The anti-CD73 antibody or antigen-binding portion of any one of claims 1-11, wherein the antibody or antigen-binding portion has at least one property selected from:
- 20 a) inhibits the activity of soluble CD73 *in vitro*;
- b) inhibits the activity of CD73 on Calu-6 cells *in vitro*;
- c) inhibits the activity of CD73 on H292 cells *in vitro*;
- 25 d) specifically binds to human and cynomolgus CD73 expressed on CHO-S cells;
- e) binds to the ECD of human CD73 with a  $K_D$  of 1 nM or less as measured by SPR;
- f) binds to the ECD of cynomolgus CD73 with a  $K_D$  of 0.7 nM or less as measured by SPR;
- 30 g) does not bind to the same epitope of CD73 as oleclumab, CPX006, and/or 11E1;

- h) binds an epitope on the CD73 homodimer in a manner that gives rise to a 1:1 complex;
  - i) inhibits soluble CD73 activity more effectively than oleclumab *in vitro*;
  - j) inhibits CD73 activity on Calu-6, H292, and Cynom-K1 cells *in vitro*;
  - 5 k) inhibits CD73 activity on Calu-6, NCI-H1775, KYSE-30, and Capan-2 cells *in vitro*;
  - l) inhibits proliferation of MDA-MB-231 and MDA-MB-468 cells *in vitro*;
  - m) inhibits CD73 activity on primary CD4<sup>+</sup> and CD8<sup>+</sup> T cells and CD19<sup>+</sup> B cells *in vitro*;
  - 10 n) restores proliferation of CD4<sup>+</sup> T cells *in vitro*;
  - o) activates CD4<sup>+</sup> and CD8<sup>+</sup> T cells *in vitro*;
  - p) in combination with an anti-PD-1 antibody, restores T cell proliferation in a one-way mixed lymphocyte reaction (MLR) in the presence of AMP;
  - 15 q) enhances T cell activation in the presence of AMP in combination with an anti-PD-1 antibody, in a one-way MLR;
  - r) does not stimulate B cell activation *in vitro*;
  - s) does not reduce levels of CD73 in H292 cells *in vitro* by more than 25%;
  - 20 t) inhibits CD73 activity in tumors harvested from PBMC-humanized mice engrafted with A375 cells;
  - u) inhibits tumor growth *in vivo* in NOD-scid mice engrafted with MDA-MB-231 cells;
  - v) inhibits tumor growth *in vivo* in PBMC-humanized mice engrafted with Calu-6 cells; and
  - 25 w) inhibits tumor growth *in vivo* in PBMC-humanized mice engrafted with A375 cells.
13. The anti-CD73 antibody or antigen-binding portion of claim 12, wherein the antibody or antigen-binding portion has at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 11,
- 30 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, or all 23 of said properties.
14. The anti-CD73 antibody of any one of claims 1-13, wherein the antibody is an IgG.

15. The anti-CD73 antibody of claim 14, wherein the antibody is an IgG<sub>1</sub>.
16. The anti-CD73 antibody of any one of claims 1-15, wherein the antibody comprises at least one mutation in the F<sub>C</sub> region.
- 5 17. The anti-CD73 antibody of any one of claims 1-16, wherein the antibody is an IgG<sub>1</sub> and comprises a mutation in one or more of heavy chain amino acid positions 234 and 235, which are numbered according to the IMGT® numbering scheme.
- 10 18. The anti-CD73 antibody of claim 17, wherein one or both of the amino acid residues at positions 234 and 235 are mutated from Leu to Ala.
19. A pharmaceutical composition comprising an anti-CD73 antibody or antigen-binding portion thereof of any one of claims 1-18 and a pharmaceutically acceptable excipient.
- 15 20. The pharmaceutical composition of claim 19, further comprising an immunostimulatory agent, a vaccine, a chemotherapeutic agent, an anti-neoplastic agent, an anti-angiogenic agent, a tyrosine kinase inhibitor, or a CD73 pathway inhibitor.
- 20 21. An isolated nucleic acid molecule comprising a nucleotide sequence that encodes the heavy chain or an antigen-binding portion thereof, or a nucleotide sequence that encodes the light chain or an antigen-binding portion thereof, or both, of the anti-CD73 antibody of any one of claims 1-18.
22. The isolated nucleic acid molecule of claim 21, wherein said nucleic acid molecule comprises the nucleotide sequence of any one of SEQ ID NOs: 1-8.
- 25 23. A vector comprising the isolated nucleic acid molecule of claim 21 or 22, wherein said vector further comprises an expression control sequence.

24. A host cell comprising a nucleotide sequence that encodes the heavy chain or an antigen-binding portion thereof, and a nucleotide sequence that encodes the light chain or an antigen-binding portion thereof, of the anti-CD73 antibody of any one of claims 1-18.
- 5 25. A method for producing an anti-CD73 antibody or an antigen-binding portion thereof, comprising providing a host cell according to claim 24, culturing said host cell under conditions suitable for expression of the antibody or portion, and isolating the resulting antibody or portion.
- 10 26. A bi-specific binding molecule comprising the antigen-binding portion of one or two distinct anti-CD73 antibodies according to any one of claims 1-18.
- 15 27. A method for decreasing CD73 activity in a patient in need thereof, comprising administering to said patient a therapeutically effective amount of the anti-CD73 antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical composition of claim 19 or 20, or the bi-specific binding molecule of claim 26.
- 20 28. A method for increasing CD4<sup>+</sup> T cell proliferation in a patient in need thereof, comprising administering to said patient a therapeutically effective amount of the anti-CD73 antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical composition of claim 19 or 20, or the bi-specific binding molecule of claim 26.
- 25 29. A method for stimulating the immune system in a patient in need thereof, comprising administering to said patient a therapeutically effective amount of the anti-CD73 antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical composition of claim 19 or 20, or the bi-specific binding molecule of claim 26.
30. A method for treating cancer in a patient, comprising administering to said patient a therapeutically effective amount of the anti-CD73 antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical

composition of claim 19 or 20, or the bi-specific binding molecule of claim 26.

31. The method of claim 30, wherein the cancer originates in a tissue selected from the group consisting of skin, lung, intestine, colon, ovary, brain, prostate, kidney, soft tissues, the hematopoietic system, head and neck, liver, bone, bladder, breast, stomach, uterus, cervix, and pancreas.
32. The method of claim 30, wherein the cancer is melanoma, head and neck cancer, breast cancer, bladder cancer, non-small cell lung cancer, pancreatic cancer, ovarian cancer, renal cell carcinoma, prostate cancer, colorectal cancer, cholangiocarcinoma, thyroid cancer, or testicular cancer.
33. The method of any one of claims 27-32, further comprising administering to the patient an immunostimulatory agent, a vaccine, a chemotherapeutic agent, an anti-neoplastic agent, an anti-angiogenic agent, a tyrosine kinase inhibitor, a CD73 pathway inhibitor, or radiation therapy.
34. Use of the anti-CD73 antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical composition of claim 19 or 20, or the bi-specific binding molecule of claim 26 for the manufacture of a medicament for:
- a) decreasing CD73 activity in a patient;
  - b) increasing CD4<sup>+</sup> T cell proliferation in a patient;
  - c) stimulating the immune system in a patient; or
  - d) treating cancer in a patient.
35. The antibody or antigen-binding portion of any one of claims 1-18, the pharmaceutical composition of claim 19 or 20, or the bi-specific binding molecule of claim 26 for use in:
- a) decreasing CD73 activity in a patient;
  - b) increasing CD4<sup>+</sup> T cell proliferation in a patient;
  - c) stimulating the immune system in a patient; or
  - d) treating cancer in a patient.