

CLAIMS

1. An isolated antibody or antigen-binding fragment thereof, comprising: (a) an immunoglobulin heavy chain variable region that comprises a heavy chain complementarity determining region 1 (VH CDR1) comprising the amino acid sequence set forth in SEQ ID NO: 2 or a variant thereof, wherein the variant consists of a Y→A substitution at position 33 of SEQ ID NO:35; a heavy chain complementarity determining region 2 (VH CDR2) comprising the amino acid sequence set forth in SEQ ID NO: 3 or a variant thereof, wherein the variant consists of a T→S substitution at position 55 of SEQ ID NO:35; and a heavy chain complementarity determining region 3 (VH CDR3) comprising the amino acid sequence set forth in SEQ ID NO: 4 or a variant thereof, wherein the variant consists of a Y→A substitution at position 104 or an A→H substitution at position 106 of SEQ ID NO:35; and (b) an immunoglobulin light chain variable region that comprises a light chain complementarity determining region 1 (VL CDR1) comprising the amino acid sequence set forth in SEQ ID NO: 6 or a variant thereof, wherein the variant consists of a Y→A substitution at position 30 of SEQ ID NO:5; a light chain complementarity determining region 2 (VL CDR2) comprising the amino acid sequence set forth in SEQ ID NO: 7 or a variant thereof, wherein the variant consists of G→A substitution at position 50 of SEQ ID NO:5; and a light chain complementarity determining region 3 (VL CDR3) comprising the amino acid sequence set forth in SEQ ID NO: 8 or a variant thereof, wherein the variant consists of a T→S substitution at position 93 of SEQ ID NO:5;

wherein the VH comprises an amino acid sequence that has at least 90 percent identity to the amino acid sequence set forth in SEQ ID NO:35 and the VL comprises an amino acid sequence that has at least 90 percent identity to the amino acid sequence set forth in SEQ ID NO:5,

wherein the antibody or antigen-binding fragment thereof is capable of binding specifically:

to a biantennary Le^B/Le^B antigen comprising Fuc₄(Galβ1→3GlcNAc)₂ **[I]** or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc]₂ **[III]**,

to a biantennary Le^Y/Le^Y antigen comprising Fuc₄(Galβ1→4GlcNAc)₂ **[III]** or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc]₂ **[IV]**,

to a biantennary Le^B/Le^Y antigen comprising Fuc₂(Galβ1→3GlcNAc)[Fuc₂(Galβ1→4GlcNAc)] **[V]**, or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc][Fucα1-2Galβ1-4(Fucα1-3)GlcNAc] **[VI]**,

and to a biantennary Le^Y/Le^B antigen comprising Fuc₂(Galβ1-4GlcNAc)[Fuc₂(Galβ1-3GlcNAc)] **[VII]** or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc][Fucα1-2Galβ1-3(Fucα1-4)GlcNAc] **[VIII]**,

and wherein the antibody or antigen-binding fragment thereof does not bind specifically to a monoantennary Le^x antigen that comprises Galβ1→4(Fucα1→3)GlcNAc **[IX]**, or to a biantennary Le^x antigen that comprises [Galβ1→4(Fucα1→3)GlcNAc]₂ **[X]**, or to a monoantennary Le^A antigen that comprises Galβ1-3(Fucα1-4)GlcNAc **[XI]**, or to a monoantennary H antigen type 2 that comprises Fucα1-2Galβ1-4GlcNAc **[XII]**, or to a biantennary H antigen type 2 that comprises (Fucα1-2Galβ1-4GlcNAc)₂ **[XIII]** or to a monoantennary H antigen type 1 that comprises Fucα1-2Galβ1-3GlcNAc **[XIV]**.

2. An isolated antibody or antigen-binding fragment thereof, comprising: (a) an immunoglobulin heavy chain variable region that comprises a heavy chain complementarity determining region 1 (VH CDR1) comprising the amino acid sequence set forth in SEQ ID NO: 2; a heavy chain complementarity determining region 2 (VH CDR2) comprising the amino acid sequence set forth in SEQ ID NO: 3; a heavy chain complementarity determining region 3 (VH CDR3) comprising the amino acid sequence set forth in SEQ ID NO: 4; and (b) an immunoglobulin light chain variable region that comprises a light chain complementarity determining region 1 (VL CDR1) comprising the amino acid sequence set forth in SEQ ID NO: 6; a light chain

complementarity determining region 2 (VL CDR2) comprising the amino acid sequence set forth in SEQ ID NO: 7; and a light chain complementarity determining region 3 (VL CDR3) comprising the amino acid sequence set forth in SEQ ID NO: 8; wherein the antibody or antigen-binding fragment thereof is capable of binding specifically:

to a biantennary Le^B/Le^B antigen comprising Fuc₄(Galβ1→3GlcNAc)₂ [I] or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc]₂ [III],

to a biantennary Le^Y/Le^Y antigen comprising Fuc₄(Galβ1→4GlcNAc)₂ [III] or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc]₂ [IV],

to a biantennary Le^B/Le^Y antigen comprising Fuc₂(Galβ1→3GlcNAc)[Fuc₂(Galβ1→4GlcNAc)] [V], or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc][Fucα1-2Galβ1-4(Fucα1-3)GlcNAc] [VI],

and to a biantennary Le^Y/Le^B antigen comprising Fuc₂(Galβ1-4GlcNAc)[Fuc₂(Galβ1-3GlcNAc)] [VII] or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc][Fucα1-2Galβ1-3(Fucα1-4)GlcNAc] [VIII],

and wherein the antibody or antigen-binding fragment thereof does not bind specifically to a monoantennary Le^x antigen that comprises Galβ1→4(Fucα1→3)GlcNAc [IX], or to a biantennary Le^x antigen that comprises [Galβ1→4(Fucα1→3)GlcNAc]₂ [X], or to a monoantennary Le^A antigen that comprises Galβ1-3(Fucα1-4)GlcNAc [XI], or to a monoantennary H antigen type 2 that comprises Fucα1-2Galβ1-4GlcNAc [XII], or to a biantennary H antigen type 2 that comprises (Fucα1-2Galβ1-4GlcNAc)₂ [XIII] or to a monoantennary H antigen type 1 that comprises Fucα1-2Galβ1-3GlcNAc [XIV].

3. An isolated antibody or antigen-binding fragment thereof, comprising: an immunoglobulin heavy chain variable region and an immunoglobulin light chain variable region, wherein the immunoglobulin heavy chain variable region and the immunoglobulin light chain variable region respectively comprise:

(i) a heavy chain complementarity determining region 1 (VH CDR1) comprising a variant of SEQ ID NO: 2, wherein the variant consists of a Y→A substitution at position 33 of SEQ ID NO: 35; a heavy chain complementarity

determining region 2 (VH CDR2) comprising SEQ ID NO: 3; and a heavy chain complementarity determining region 3 (VH CDR3) comprising SEQ ID NO: 4; and a light chain complementarity determining region 1 (VL CDR1) comprising SEQ ID NO: 6; a light chain complementarity determining region 2 (VL CDR2) comprising SEQ ID NO: 7; and a light chain complementarity determining region 3 (VL CDR3) comprising SEQ ID NO: 8;

(ii) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising a variant of SEQ ID NO: 3, wherein the variant consists of a T→S substitution at position 55 of SEQ ID NO.:35; and a VH CDR3 comprising SEQ ID NO: 4; and

a VL CDR1 comprising SEQ ID NO: 6; a VL CDR2 comprising SEQ ID NO: 7; and a VL CDR3 comprising SEQ ID NO: 8;

(iii) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising SEQ ID NO: 3; and a VH CDR3 comprising a variant of SEQ ID NO: 4, wherein the variant consists of a Y→A substitution at position 104 of SEQ ID NO: 35, or a A→H substitution at position 106 of SEQ ID NO: 35; and

a VL CDR1 comprising SEQ ID NO: 6; a VL CDR2 comprising SEQ ID NO: 7; and a VL CDR3 comprising SEQ ID NO: 8;

(iv) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising SEQ ID NO: 3; and a VH CDR3 comprising SEQ ID NO: 4; and

a VL CDR1 comprising a variant of SEQ ID NO: 6, wherein the variant consists of a Y→A substitution at position 30 of SEQ ID NO: 5; a VL CDR2 comprising SEQ ID NO: 7; and a VL CDR3 comprising SEQ ID NO: 8;

(v) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising SEQ ID NO: 3; and a VH CDR3 comprising SEQ ID NO: 4; and

a VL CDR1 comprising SEQ ID NO: 6; a VL CDR2 comprising a variant of SEQ ID NO: 7, wherein the variant consists of a G→A substitution at position 50 of SEQ ID NO: 5; and a VL CDR3 comprising SEQ ID NO: 8;

(vi) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising SEQ ID NO: 3; and a VH CDR3 comprising SEQ ID NO: 4; and a VL CDR1 comprising SEQ ID NO: 6; a VL CDR2 comprising SEQ ID NO: 7; and a VL CDR3 comprising a

variant of SEQ ID NO: 8, wherein the variant consists of a T→S substitution at position 93 of SEQ ID NO: 5; or

(vii) a VH CDR1 comprising SEQ ID NO: 2; a VH CDR2 comprising a variant of SEQ ID NO: 3, wherein the variant consists of a T→S substitution at position 55 of SEQ ID NO: 35; and a VH CDR3 comprising a variant of SEQ ID NO: 4, wherein the variant consists of a Y→A substitution at position 104 of SEQ ID NO: 35, or a A→H substitution at position 106 of SEQ ID NO: 35; and

a VL CDR1 comprising SEQ ID NO: 6; a VL CDR2 comprising SEQ ID NO: 7; and a VL CDR3 comprising SEQ ID NO: 8,

wherein the VH comprises an amino acid sequence that has at least 90 percent identity to the amino acid sequence set forth in SEQ ID NO:35 and the VL comprises an amino acid sequence that has at least 90 percent identity to the amino acid sequence set forth in SEQ ID NO:5,

wherein the antibody or antigen-binding fragment thereof is capable of binding specifically:

to a biantennary Le^B/Le^B antigen comprising Fuc₄(Galβ1→3GlcNAc)₂ **[I]** or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc]₂ **[III]**,

to a biantennary Le^Y/Le^Y antigen comprising Fuc₄(Galβ1→4GlcNAc)₂ **[III]** or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc]₂ **[IV]**,

to a biantennary Le^B/Le^Y antigen comprising Fuc₂(Galβ1→3GlcNAc)[Fuc₂(Galβ1→4GlcNAc)] **[V]**, or [Fucα1-2Galβ1-3(Fucα1-4)GlcNAc][Fucα1-2Galβ1-4(Fucα1-3)GlcNAc] **[VI]**,

and to a biantennary Le^Y/Le^B antigen comprising Fuc₂(Galβ1-4GlcNAc)[Fuc₂(Galβ1-3GlcNAc)] **[VII]**, or [Fucα1-2Galβ1-4(Fucα1-3)GlcNAc][Fucα1-2Galβ1-3(Fucα1-4)GlcNAc] **[VIII]**,

and wherein the antibody or antigen-binding fragment thereof does not bind specifically to a monoantennary Le^x antigen that comprises Galβ1→4(Fucα1→3)GlcNAc **[IX]**, or to a biantennary Le^x antigen that comprises [Galβ1→4(Fucα1→3)GlcNAc]₂ **[X]**, or to a monoantennary Le^A antigen that comprises

Gal β 1-3(Fuc α 1-4)GlcNAc **[XI]**, or to a monoantennary H antigen type 2 that comprises Fuc α 1-2Gal β 1-4GlcNAc **[XII]**, or to a biantennary H antigen type 2 that comprises (Fuc α 1-2Gal β 1-4GlcNAc)₂ **[XIII]** or to a monoantennary H antigen type 1 that comprises Fuc α 1-2Gal β 1-3GlcNAc **[XIV]**.

4. An isolated antibody or antigen-binding fragment thereof, comprising: an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 35; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 5, wherein the antibody or antigen-binding fragment thereof is capable of binding specifically:

to a biantennary Le^B/Le^B antigen comprising Fuc₄(Gal β 1→3GlcNAc)₂ **[I]** or [Fuc α 1-2Gal β 1-3(Fuc α 1-4)GlcNAc]₂ **[III]**,

to a biantennary Le^Y/Le^Y antigen comprising Fuc₄(Gal β 1→4GlcNAc)₂ **[III]** or [Fuc α 1-2Gal β 1-4(Fuc α 1-3)GlcNAc]₂ **[IV]**,

to a biantennary Le^B/Le^Y antigen comprising Fuc₂(Gal β 1→3GlcNAc)[Fuc₂(Gal β 1→4GlcNAc)] **[V]**, or [Fuc α 1-2Gal β 1-3(Fuc α 1-4)GlcNAc][Fuc α 1-2Gal β 1-4(Fuc α 1-3)GlcNAc] **[VI]**,

and to a biantennary Le^Y/Le^B antigen comprising Fuc₂(Gal β 1-4GlcNAc)[Fuc₂(Gal β 1-3GlcNAc)] **[VII]** or [Fuc α 1-2Gal β 1-4(Fuc α 1-3)GlcNAc][Fuc α 1-2Gal β 1-3(Fuc α 1-4)GlcNAc] **[VIII]**,

and wherein the antibody or antigen-binding fragment thereof does not bind specifically to a monoantennary Le^x antigen that comprises Gal β 1→4(Fuc α 1→3)GlcNAc **[IX]**, or to a biantennary Le^x antigen that comprises [Gal β 1→4(Fuc α 1→3)GlcNAc]₂ **[X]**, or to a monoantennary Le^A antigen that comprises Gal β 1-3(Fuc α 1-4)GlcNAc **[XI]**, or to a monoantennary H antigen type 2 that comprises Fuc α 1-2Gal β 1-4GlcNAc **[XII]**, or to a biantennary H antigen type 2 that comprises (Fuc α 1-2Gal β 1-4GlcNAc)₂ **[XIII]** or to a monoantennary H antigen type 1 that comprises Fuc α 1-2Gal β 1-3GlcNAc **[XIV]**.

5. An isolated antibody or antigen-binding fragment thereof, comprising:

(1) an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 32; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 31;

(2) an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 32; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 33;

(3) an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 34; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 33;

(4) an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 32; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 5; or

(5) an immunoglobulin heavy chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 1; and an immunoglobulin light chain variable region that comprises the amino acid sequence set forth in SEQ ID NO: 5,

wherein the antibody or antigen-binding fragment thereof is capable of binding specifically:

to a biantennary Le^B/Le^B antigen comprising $\text{Fuc}_4(\text{Gal}\beta 1 \rightarrow 3\text{GlcNAc})_2$ **[I]** or $[\text{Fuc}\alpha 1-2\text{Gal}\beta 1-3(\text{Fuc}\alpha 1-4)\text{GlcNAc}]_2$ **[III]**,

to a biantennary Le^Y/Le^Y antigen comprising $\text{Fuc}_4(\text{Gal}\beta 1 \rightarrow 4\text{GlcNAc})_2$ **[III]** or $[\text{Fuc}\alpha 1-2\text{Gal}\beta 1-4(\text{Fuc}\alpha 1-3)\text{GlcNAc}]_2$ **[IV]**,

to a biantennary Le^B/Le^Y antigen comprising $\text{Fuc}_2(\text{Gal}\beta 1 \rightarrow 3\text{GlcNAc})[\text{Fuc}_2(\text{Gal}\beta 1 \rightarrow 4\text{GlcNAc})]$ **[V]**, or $[\text{Fuc}\alpha 1-2\text{Gal}\beta 1-3(\text{Fuc}\alpha 1-4)\text{GlcNAc}][\text{Fuc}\alpha 1-2\text{Gal}\beta 1-4(\text{Fuc}\alpha 1-3)\text{GlcNAc}]$ **[VI]**,

and to a biantennary Le^Y/Le^B antigen comprising $\text{Fuc}_2(\text{Gal}\beta 1-4\text{GlcNAc})[\text{Fuc}_2(\text{Gal}\beta 1-3\text{GlcNAc})]$ **[VII]** or $[\text{Fuc}\alpha 1-2\text{Gal}\beta 1-4(\text{Fuc}\alpha 1-3)\text{GlcNAc}][\text{Fuc}\alpha 1-2\text{Gal}\beta 1-3(\text{Fuc}\alpha 1-4)\text{GlcNAc}]$ **[VIII]**,

and wherein the antibody or antigen-binding fragment thereof does not bind specifically to a monoantennary Le^x antigen that comprises Galβ1→4(Fucα1→3)GlcNAc **[IX]**, or to a biantennary Le^x antigen that comprises [Galβ1→4(Fucα1→3)GlcNAc]₂ **[X]**, or to a monoantennary Le^A antigen that comprises Galβ1-3(Fucα1-4)GlcNAc **[XI]**, or to a monoantennary H antigen type 2 that comprises Fucα1-2Galβ1-4GlcNAc **[XII]**, or to a biantennary H antigen type 2 that comprises (Fucα1-2Galβ1-4GlcNAc)₂ **[XIII]** or to a monoantennary H antigen type 1 that comprises Fucα1-2Galβ1-3GlcNAc **[XIV]**.

6. An isolated antibody comprising an immunoglobulin heavy chain having the amino acid sequence set forth in SEQ ID NO: 10 and an immunoglobulin light chain having the amino acid sequence set forth in SEQ ID NO: 11.

7. The isolated antibody of claim 6, which is monoclonal.

8. The isolated antibody or antigen-binding fragment thereof of claim 2 or 4, wherein the antibody is monoclonal.

9. The isolated antibody or antigen-binding fragment thereof of claim 2 or 4, wherein the antibody is a humanized antibody.

10. The isolated antibody or antigen-binding fragment thereof of claim 2 or 4, wherein the antigen-binding fragment is selected from a Fab fragment, an F(ab')₂ fragment, an Fv fragment, a single chain Fv (ScFv) antibody, and a diabody.

11. A pharmaceutical composition comprising: the isolated antibody or an antigen-binding fragment thereof of claim 2, 4, or 6; and a pharmaceutical carrier.

12. The antibody or antigen-binding fragment of claim 2, wherein the heavy chain variable region comprises an amino acid sequence that has at least 90

percent identity to the amino acid sequence set forth in SEQ ID NO: 35, and the immunoglobulin light chain variable region comprises an amino acid sequence that has at least 90 percent identity to the amino acid sequence set forth in SEQ ID NO: 5.

13. The antibody or antigen-binding fragment of claim 2, wherein the heavy chain variable region comprises an amino acid sequence that has at least 95 percent identity to the amino acid sequence set forth in SEQ ID NO: 35, and the immunoglobulin light chain variable region comprises an amino acid sequence that has at least 95 percent identity to the amino acid sequence set forth in SEQ ID NO: 5.

14. The antibody or antigen-binding fragment of claim 2, wherein the heavy chain variable region comprises an amino acid sequence that has at least 97 percent identity to the amino acid sequence set forth in SEQ ID NO: 35, and the immunoglobulin light chain variable region comprises an amino acid sequence that has at least 97 percent identity to the amino acid sequence set forth in SEQ ID NO: 5.

15. Use of the isolated antibody or antigen-binding fragment of any one of claims 1-10 and 12-14, in the manufacture of a medicament for treating a cancer that expresses one or more biantennary Lewis antigens selected from the group consisting of biantennary Le^B/Le^B, biantennary Le^Y/Le^Y, biantennary Le^B/Le^Y, and biantennary Le^Y/Le^B.

16. Use of the isolated antibody or antigen-binding fragment of any one of claims 1-10 and 12-14, in the manufacture of a medicament for treating colorectal adenocarcinoma, gastric adenocarcinoma or lung cancer that expresses one or more biantennary Lewis antigens selected from the group consisting of biantennary Le^B/Le^B, biantennary Le^Y/Le^Y, biantennary Le^B/Le^Y, and biantennary Le^Y/Le^B.

17. Use of the pharmaceutical composition of claim 11, in the manufacture of a medicament for treating a cancer that expresses one or more

biantennary Lewis antigens selected from the group consisting of biantennary Le^B/Le^B, biantennary Le^Y/Le^Y, biantennary Le^B/Le^Y, and biantennary Le^Y/Le^B.

18. Use of the pharmaceutical composition of claim 11, in the manufacture of a medicament for treating colorectal adenocarcinoma, gastric adenocarcinoma or lung cancer that expresses one or more biantennary Lewis antigens selected from the group consisting of biantennary Le^B/Le^B, biantennary Le^Y/Le^Y, biantennary Le^B/Le^Y, and biantennary Le^Y/Le^B.