

WE CLAIMS:

1. An antibody or antigen-binding fragment thereof that:
  - (a) binds an epitope within amino acids 101 to 346 of human CD123 / IL3-R $\alpha$  antigen, and
  - (b) inhibits IL3-dependent proliferation in antigen-positive TF-1 cells.
- ~~2. The antibody or antigen-binding fragment thereof of claim 1, wherein said antibody or antigen-binding fragment thereof binds an epitope within amino acids 101 to 204 of human CD123 antigen.~~
- ~~3. The antibody or antigen-binding fragment thereof of claim 1, wherein said antibody or antigen-binding fragment thereof binds an epitope within amino acids 205 to 346 of human CD123 antigen.~~
- 2 4. An antibody or antigen-binding fragment thereof that:
  - (a) binds an epitope within amino acids 1 to 100 of human CD123 antigen, and
  - (b) inhibits IL3-dependent proliferation in antigen-positive TF-1 cells, with an IC<sub>50</sub> value of 0.1 nM or less (e.g., 0.08 nM, 0.05 nM, 0.03 nM).
- ~~5. The antibody or antigen-binding fragment thereof of any of claims 1-4, wherein said antibody or antigen-binding fragment thereof inhibits the proliferation of leukemic stem cells or leukemic blast cells but not hematopoietic stem cells.~~
- ~~6. The antibody or antigen-binding fragment thereof of any of claims 1-5, wherein said antibody or antigen-binding fragment thereof binds to human CD123 antigen-positive cells with a dissociation constant (K<sub>d</sub>) of 0.3 nM or lower.~~
- ~~7. The antibody or antigen-binding fragment thereof of claim 6, wherein said antibody or antigen-binding fragment thereof binds to human CD123 antigen-positive cells with a K<sub>d</sub> of between 0.01 nM and 0.3 nM.~~
- ~~8. The antibody or antigen-binding fragment thereof of claim 7, wherein said antibody or antigen-binding fragment thereof binds to human CD123 antigen-positive cells with a K<sub>d</sub> of between 0.01 nM and 0.2 nM.~~
- ~~9. The antibody or antigen-binding fragment thereof of claim 8, wherein said antibody or~~

~~antigen-binding fragment thereof binds to human CD123 antigen-positive cells with a  $K_d$  of between 0.01 nM and 0.1 nM.~~

- ~~10. The antibody or antigen-binding fragment thereof of any one of claims 1-9, wherein the antibody or antigen-binding fragment thereof binds to cynomolgus monkey CD123.~~
- ~~11. The antibody or antigen-binding fragment thereof of any one of claims 1-9, wherein the antibody or antigen-binding fragment thereof binds to cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.3 nM.~~
- ~~12. The antibody or antigen-binding fragment thereof of any one of claims 1-9, wherein the antibody or antigen-binding fragment thereof binds to cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.2 nM.~~
- ~~13. The antibody or antigen-binding fragment thereof of any one of claims 1-9, wherein the antibody or antigen-binding fragment thereof binds to cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.1 nM.~~
- ~~14. The antibody or antigen-binding fragment thereof of any one of claims 1-13, wherein the antibody or antigen-binding fragment thereof binds both human and cynomolgus monkey CD123 with a substantially similar binding affinity.~~
- ~~15. The antibody or antigen-binding fragment thereof of claim 14, wherein the antibody or antigen-binding fragment thereof binds to human and cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.3 nM.~~
- ~~16. The antibody or antigen-binding fragment thereof of claim 14, wherein the antibody or antigen-binding fragment thereof binds to human and cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.2 nM.~~
- ~~17. The antibody or antigen-binding fragment thereof of claim 14, wherein the antibody or antigen-binding fragment thereof binds to human and cynomolgus monkey CD123 with a  $K_d$  of between 0.05 and 0.1 nM.~~
- ~~18. The antibody or antigen-binding fragment thereof of any one of claims 6-17, wherein  $K_d$  is measured by flow cytometry, surface plasmon resonance, or radioimmunoassay.~~
- ~~19. The antibody or antigen-binding fragment thereof of any of claims 1-18, wherein said~~

~~antibody or antigen-binding fragment thereof inhibits at least 50% of IL-3 dependent proliferation in antigen-positive TF-1 cells at a concentration of 0.5 nM or lower.~~

- 3 ~~20.~~ The antibody or antigen-binding fragment thereof of any of claims 1 and 2 ~~1-19~~, comprising:
- a) at least one heavy chain variable region or fragment thereof comprising three sequential complementarity-determining regions (CDR) CDR1, CDR2, and CDR3, respectively, wherein, with the exception of 1, 2, or 3 conservative amino acid substitutions, CDR1 is selected from the group consisting of: SEQ ID NOs: 1, 5, and 12, CDR2 is selected from the group consisting of: SEQ ID NOs: 2, 3, 6-10, 13, and 14, and, optionally, CDR3 is selected from the group consisting of: SEQ ID NOs: 4, 11, and 15; and
  - b) at least one light chain variable region or fragment thereof comprising three sequential complementarity-determining regions (CDR) CDR1, CDR2, and CDR3, respectively, wherein, with the exception of 1, 2, or 3 conservative amino acid substitutions, CDR1 is selected from the group consisting of: SEQ ID NOs: 16, 19, 20, and 23, CDR2 is selected from the group consisting of: SEQ ID NOs: 17, 21, and 24, and, optionally, CDR3 is selected from the group consisting of: SEQ ID NOs: 18, 22, and 25.
- ~~21.—The antibody or antigen-binding fragment thereof of claim 20, wherein said conservative amino acid substitutions comprise a substitution of at least one Lys in a CDR by an Arg.~~
- ~~22.—The antibody or antigen-binding fragment thereof of claim 20 or 21, wherein said antibody is a CDR-grafted humanized antibody comprising mouse CDR regions, and wherein one or more (e.g., 1, 2, 3, 4, 5, 6, 7, or 8) heavy chain and/or light chain framework region variable zone residues of said antibody is of mouse origin.~~
- ~~23.—The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- ~~a) —an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 39 or 40; and~~
  - ~~b) —an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41.~~
- 4 ~~24.~~ The antibody or antigen-binding fragment thereof of any of claims 1-3 ~~1-19~~, comprising:

- a) an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 34; and
  - b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 35.
- 5 ~~25~~. The antibody or antigen-binding fragment thereof of claim 4 ~~24~~, wherein Xaa, the second residue from the N-terminus of SEQ ID NO: 34, is Phe.
- 6 ~~26~~. The antibody or antigen-binding fragment thereof of claim 4 ~~24~~, wherein Xaa, the second residue from the N-terminus of SEQ ID NO: 34, is Val.
- ~~27. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- ~~a) an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 39 or 40, except that the N-terminal residue is Ser; and~~
  - ~~b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41.~~
- ~~28. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- ~~a) an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 39 or 40; and~~
  - ~~b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41, except that the N-terminal residue is Ser.~~
- ~~29. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- ~~a) an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 59 or 60, except that the N-terminal residue is Ser, and except that the residue corresponding to the 5<sup>th</sup> to the last residue of SEQ ID NO: 54 is Cys; and~~
  - ~~b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41.~~
- ~~30. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- ~~a) an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 59 or 60, except that the residue corresponding to the 5<sup>th</sup> to the last residue of SEQ ID NO: 54 is Cys; and~~

- b) ~~an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41, except that the N-terminal residue is Ser.~~
- ~~31. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- a) ~~an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 38; and~~
- b) ~~an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 35.~~
32. The antibody or antigen-binding fragment thereof of any of claims 1-6 ~~1-19~~, comprising:
- a) an immunoglobulin heavy chain variable region having the amino acid sequence set forth in SEQ ID NO: 34; and
- b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 37.
- ~~33. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- a) ~~an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 56; and~~
- b) ~~an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 35.~~
- ~~34. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- a) ~~an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 54; and~~
- b) ~~an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 37.~~
- ~~35. The antibody or antigen-binding fragment thereof of any one of claims 31-34, wherein Xaa, the second residue from the N-terminus of SEQ ID NOS: 38, 34, 56, and 54, is Phe.~~
- ~~36. The antibody or antigen-binding fragment thereof of any one of claims 31-34, wherein Xaa, the second residue from the N-terminus of SEQ ID NOS: 38, 34, 56, and 54, is Val.~~
- ~~37. The antibody or antigen-binding fragment thereof of any of claims 1-19, comprising:~~
- a) ~~an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 59 or 60, except that the residue corresponding to the 5<sup>th</sup> to the~~

last residue of SEQ ID NO: 54 is Cys; and

~~b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 41.~~

8 ~~38~~. The antibody or antigen-binding fragment thereof of any of claims 1-3 ~~4-19~~, comprising:

a) an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 54; and

b) an immunoglobulin light chain variable region having the amino acid sequence set forth in SEQ ID NO: 35.

9 ~~39~~. The antibody or antigen-binding fragment thereof of claim 8 ~~38, 33, or 34~~, wherein Xaa, the second residue from the N-terminus of SEQ ID NO: 54 or 56, is Phe.

10 ~~40~~. The antibody or antigen-binding fragment thereof of claim 8 ~~38, 33, or 34~~, wherein Xaa, the second residue from the N-terminus of SEQ ID NO: 54 or 56, is Val.

~~41. The antibody or antigen-binding fragment thereof of any one of claims 1-3 and 5-20, comprising:~~

~~a) an immunoglobulin heavy chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 1, a CDR2 having an amino acid sequence set forth in SEQ ID NO: 2 or 3, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 4; and~~

~~b) an immunoglobulin light chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 16, a CDR2 having an amino acid sequence set forth in SEQ ID NO: 17, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 18.~~

11 ~~42~~. The antibody or antigen-binding fragment thereof of any one of claims 1-3 ~~and 5-20~~, comprising:

a) an immunoglobulin heavy chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 5, a CDR2 having an amino acid sequence set forth in SEQ ID NO: 6, 7, 8, 9, or 10, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 11; and

b) an immunoglobulin light chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 19 or 20, a CDR2 having an amino

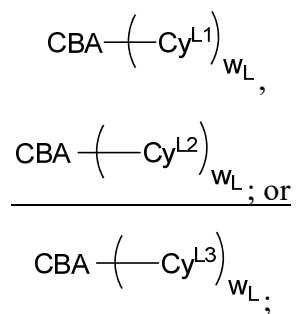
acid sequence set forth in SEQ ID NO: 21, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 22.

43. ~~The antibody or antigen-binding fragment thereof of any one of claims 4-20, comprising:~~
- a) ~~an immunoglobulin heavy chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 12, a CDR2 having an amino acid sequence set forth in SEQ ID NO: 13 or 14, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 15; and~~
  - b) ~~an immunoglobulin light chain variable region comprising a CDR1 having an amino acid sequence set forth in SEQ ID NO: 23, a CDR2 having an amino acid sequence set forth in SEQ ID NO: 24, and a CDR3 having an amino acid sequence set forth in SEQ ID NO: 25.~~
44. ~~The antibody or antigen-binding fragment thereof of any of claims 1-43, comprising:~~
- a) ~~a V<sub>H</sub> sequence at least 95% identical to a reference V<sub>H</sub> sequence selected from the group consisting of: SEQ ID NOs: 26, 28, 30, 32, 34, 38, 39, and 40 (preferably 26, 28, 30, 32, 34, and 38); and/or,~~
  - b) ~~a V<sub>L</sub> sequence at least 95% identical to a reference V<sub>L</sub> sequence selected from the group consisting of: SEQ ID NOs: 27, 29, 31, 33, 35, 37, and 41 (preferably 27, 29, 31, 35, and 37).~~
45. ~~The antibody or antigen-binding fragment thereof of claim 44, wherein the V<sub>H</sub> sequence is at least 99% identical to one of SEQ ID NOs: 26, 28, 30, 32, 34, 38, 39, and 40 (preferably 26, 28, 30, 32, 34, and 38), and/or wherein the V<sub>L</sub> sequence is at least 99% identical to one of SEQ ID NOs: 27, 29, 31, 33, 35, 37, and 41 (preferably 27, 29, 31, 35, and 37).~~
46. ~~The antibody or antigen-binding fragment thereof of claim 45, comprising:~~
- a) ~~a V<sub>H</sub> sequence selected from the group consisting of SEQ ID NOs: 26, 28, 30, 32, 34, 38, 39, and 40 (preferably 26, 28, 30, 32, 34, and 38); and/or,~~
  - b) ~~a V<sub>L</sub> sequence selected from the group consisting of SEQ ID NOs: 27, 29, 31, 33, 35, 37, and 41 (preferably 27, 29, 31, 35, and 37).~~
47. ~~The antibody or antigen-binding fragment thereof of claim 46, comprising a V<sub>H</sub> sequence of SEQ ID NO: 26 and a V<sub>L</sub> sequence of SEQ ID NO: 27.~~

- ~~48. The antibody or antigen-binding fragment thereof of claim 46, comprising a V<sub>H</sub> sequence of SEQ ID NO: 28 and a V<sub>L</sub> sequence of SEQ ID NO: 29.~~
- ~~49. The antibody or antigen-binding fragment thereof of claim 46, comprising a V<sub>H</sub> sequence of SEQ ID NO: 30 and a V<sub>L</sub> sequence of SEQ ID NO: 31.~~
- 12 ~~50.~~ The antibody or antigen-binding fragment thereof of any one of claims 1-11 ~~claim 46~~, comprising a V<sub>H</sub> sequence of SEQ ID NO: 34 and a V<sub>L</sub> sequence of SEQ ID NO: 35.
- ~~51. The antibody or antigen-binding fragment thereof of any one of claims 1-49, wherein said antibody is a murine, non-human mammal, chimeric, humanized, or human antibody.~~
- ~~52. The antibody or antigen-binding fragment thereof of claim 51, wherein said humanized antibody is a CDR-grafted antibody or resurfaced antibody.~~
- ~~53. The antibody or antigen-binding fragment thereof of any one of claims 1-52, wherein said antibody is a full-length antibody.~~
- ~~54. The antibody or antigen-binding fragment thereof of any one of claims 1-52, wherein said antigen-binding fragment thereof is an Fab, Fab', F(ab')<sub>2</sub>, F<sub>d</sub>, single chain Fv or scFv, disulfide linked F<sub>v</sub>, V-NAR domain, IgNar, intrabody, IgGΔCH<sub>2</sub>, minibody, F(ab')<sub>3</sub>, tetrabody, tribody, diabody, single domain antibody, DVD-Ig, Fcab, mAb<sub>2</sub>, (scFv)<sub>2</sub>, or scFv-Fc.~~
- 13 ~~55.~~ A polypeptide comprising the V<sub>H</sub> and V<sub>L</sub> sequences of any of claims 1-12 ~~1-54~~.
- ~~56. The polypeptide of claim 55, which is a fusion with a protein that is not a pseudomonas toxin.~~
- 14 ~~57.~~ A cell producing the antibody or antigen-binding fragment thereof of any one of claims 1-12 ~~1-54~~, or the polypeptide of claim 13 ~~55 or 56~~.
- 15 ~~58.~~ A method of producing the antibody or antigen-binding fragment thereof of any one of claims 1-12 ~~1-54~~, or the polypeptide of claim 13 ~~55 or 56~~, comprising:
- (a) ~~culturing the a cell producing said antibody or antigen-binding fragment thereof of claim 57; and,~~
  - (b) isolating said antibody, antigen-binding fragment thereof, or polypeptide from said cultured cell.

59. ~~The method of claim 58, wherein said cell is a eukaryotic cell.~~

16 60. An immunoconjugate having the following formula:

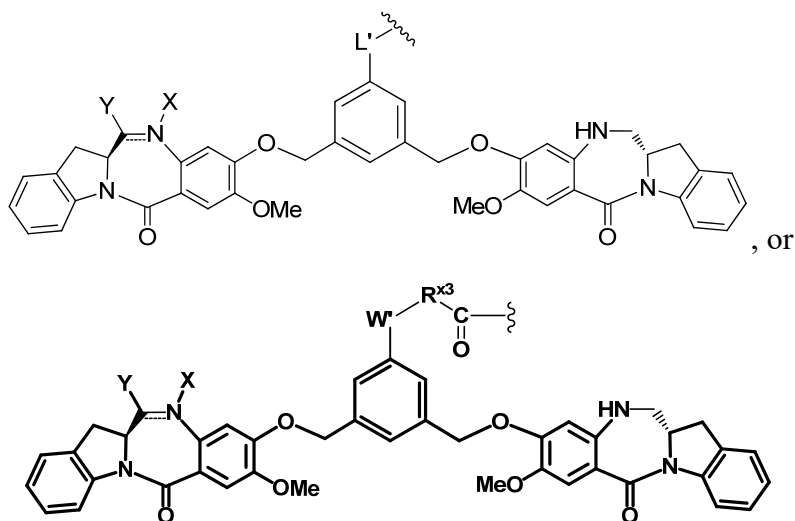


wherein:

CBA is an antibody or antigen-binding fragment thereof of any one of claims 3-6  
~~23-26~~, or the polypeptide thereof of claim 13 ~~55 or 56~~, that is covalently linked through a  
 lysine residue to Cy<sup>L1</sup>, Cy<sup>L2</sup> or Cy<sup>L3</sup>;

W<sub>L</sub> is an integer from 1 to 20; ~~and~~

Cy<sup>L1</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line == between N and C represents a single bond or a double bond,  
 provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and  
 when it is a single bond, X is -H or an amine protecting moiety, and Y is -OH or -SO<sub>3</sub>M;

W' is -NR<sup>e'</sup>,

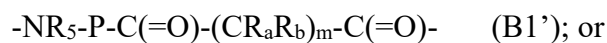
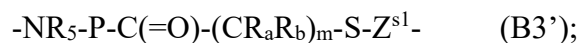
R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is -H or -Me;

R<sup>x3</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

L' is represented by the following formula:



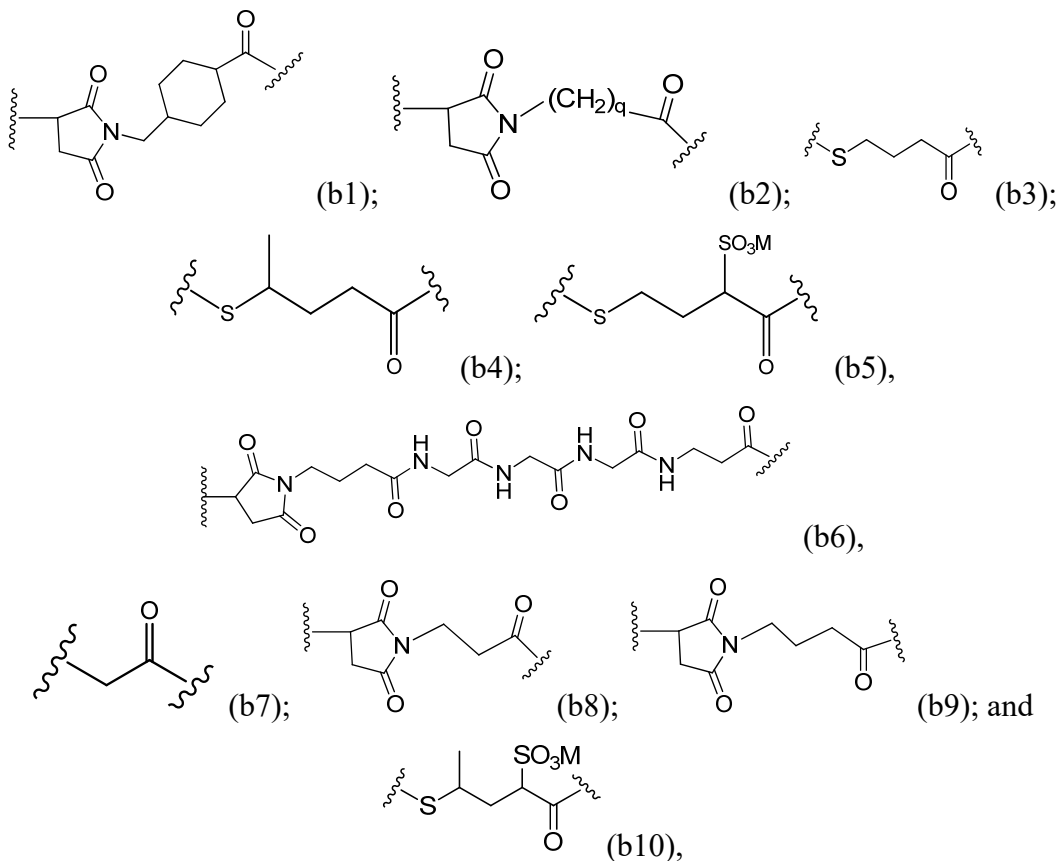
R<sub>5</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

P is an amino acid residue or a peptide containing between 2 to 20 amino acid residues;

R<sub>a</sub> and R<sub>b</sub>, for each occurrence, are each independently -H, (C<sub>1</sub>-C<sub>3</sub>)alkyl, or a charged substituent or an ionizable group Q;

m is an integer from 1 to 6; and

Z<sup>s1</sup> is selected from any one of the following formulas:

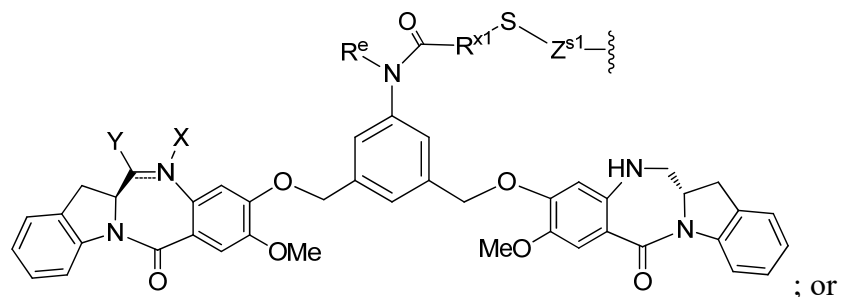
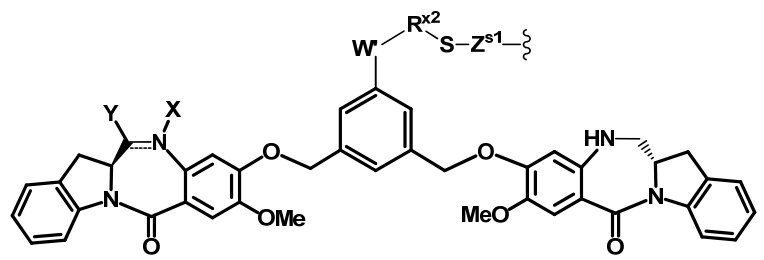


wherein:

q is an integer from 1 to 5; and

M is H<sup>+</sup> or a cation;

Cy<sup>L2</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line == between N and C represents a single bond or a double bond,  
provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and  
when it is a single bond, X is -H or an amine protecting moiety, and Y is -OH or -SO<sub>3</sub>M;

R<sup>x1</sup> and R<sup>x2</sup> are independently (C<sub>1</sub>-C<sub>6</sub>)alkyl;

R<sup>e</sup> is -H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

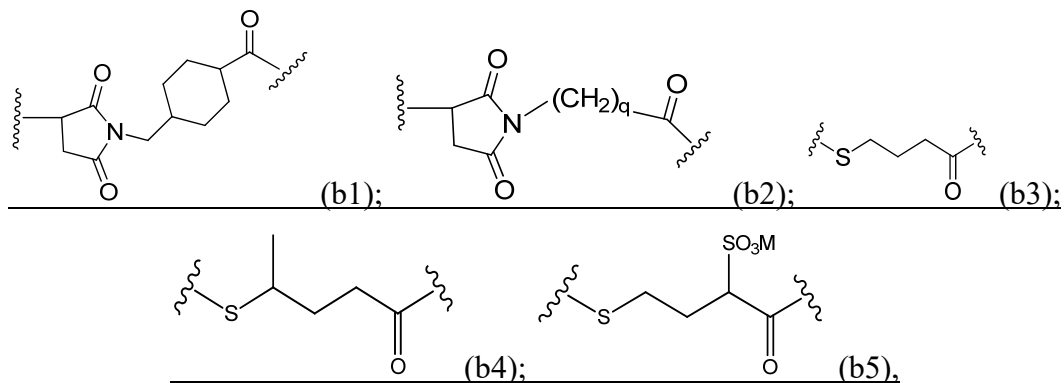
W' is -NR<sup>e'</sup>,

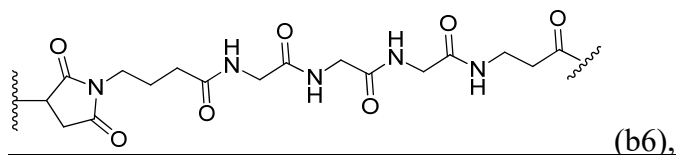
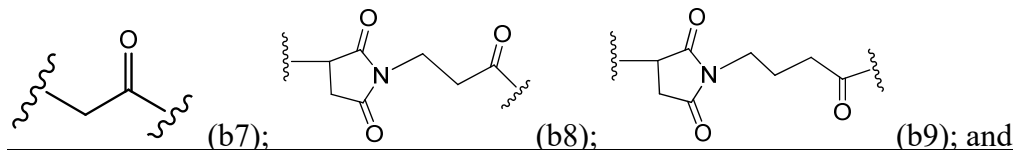
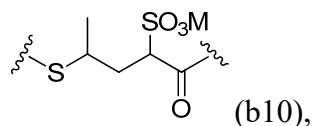
R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is -H or -Me;

Z<sup>s1</sup> is selected from any one of the following formulas:



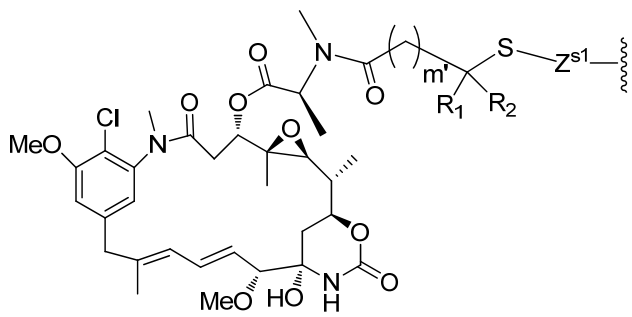


wherein:

q is an integer from 1 to 5; and

M is  $-H^+$  or a cation; and

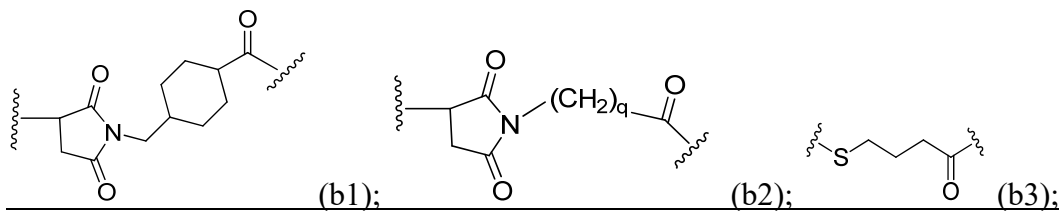
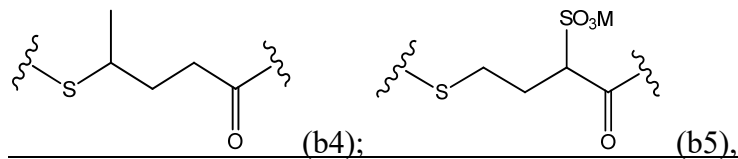
Cy<sup>L3</sup> is represented by the following formula:

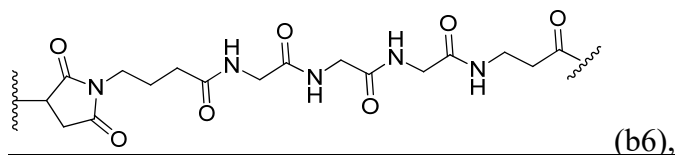
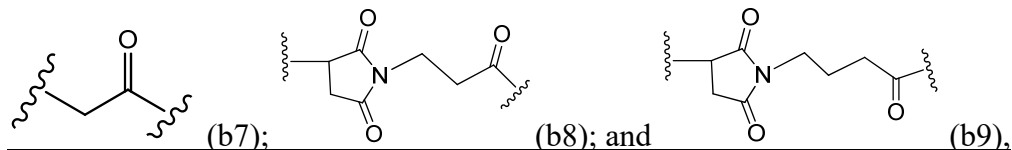


m' is 1 or 2;

R<sub>1</sub> and R<sub>2</sub>, are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

Z<sup>s1</sup> is selected from any one of the following formulas:



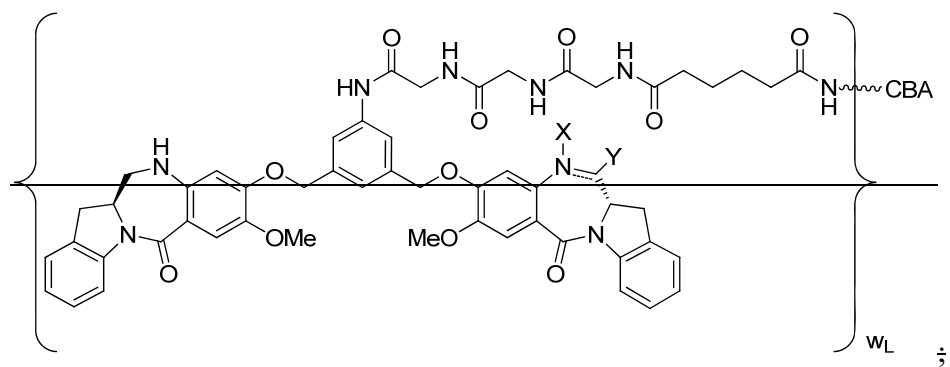
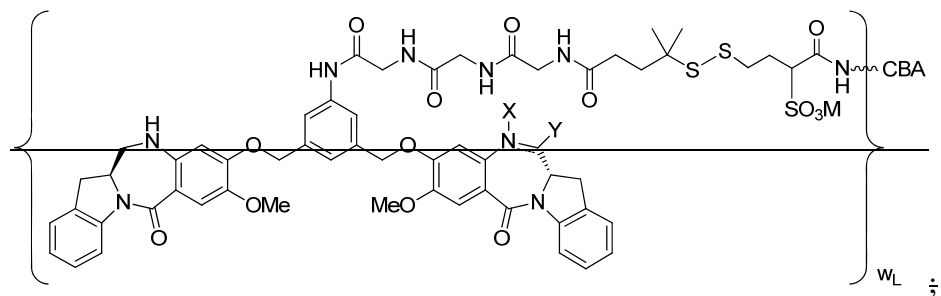
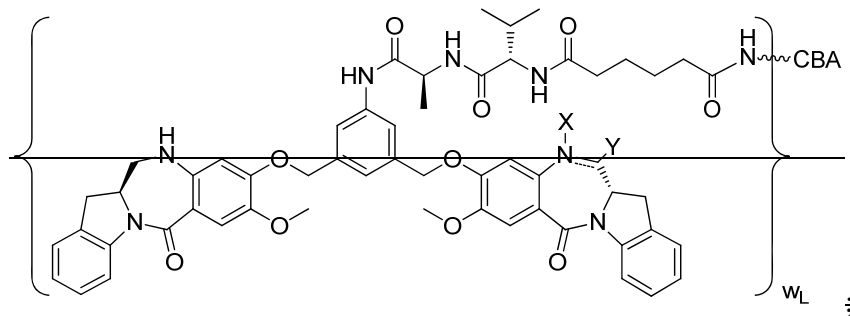
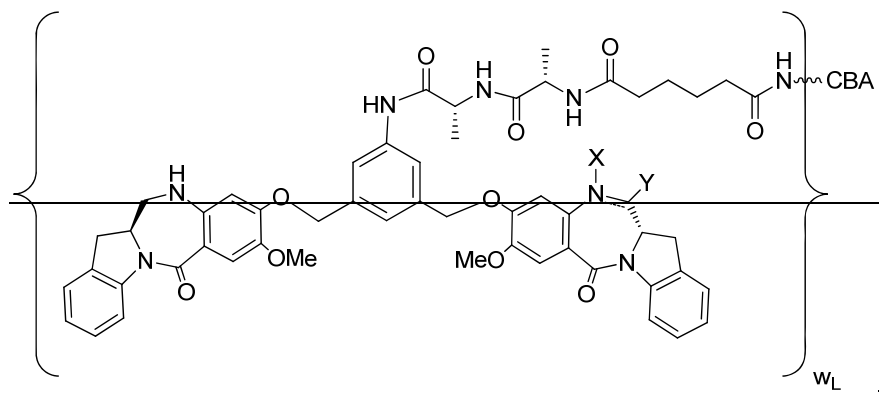


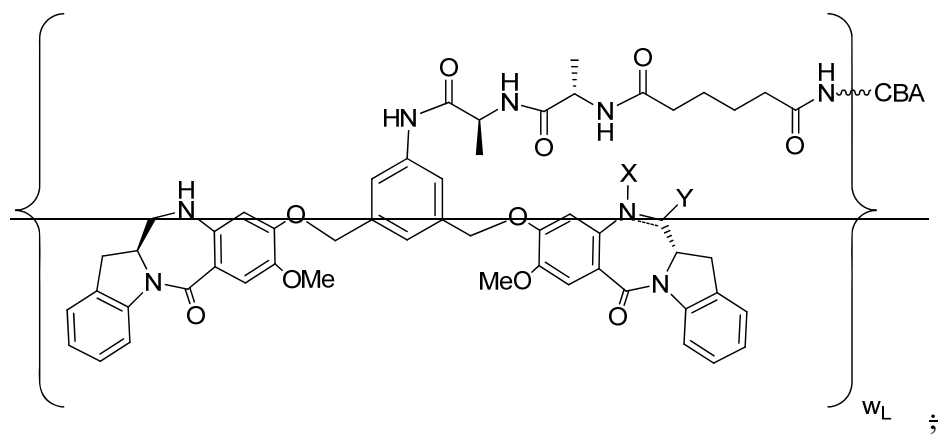
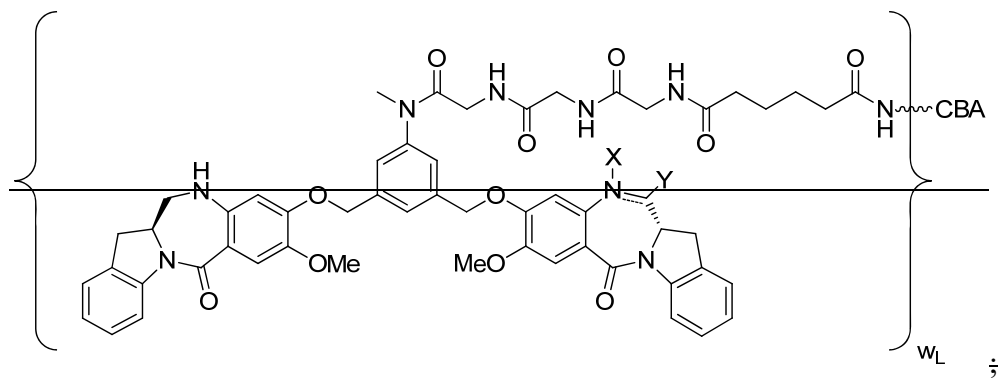
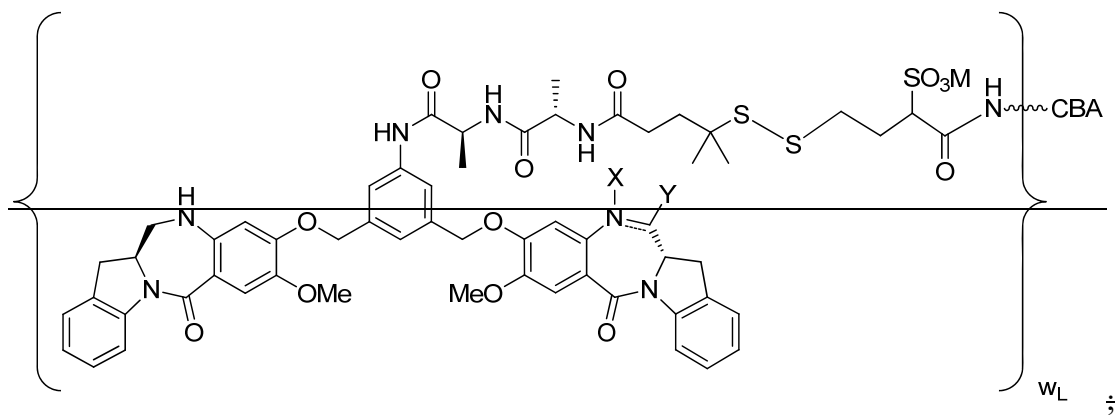
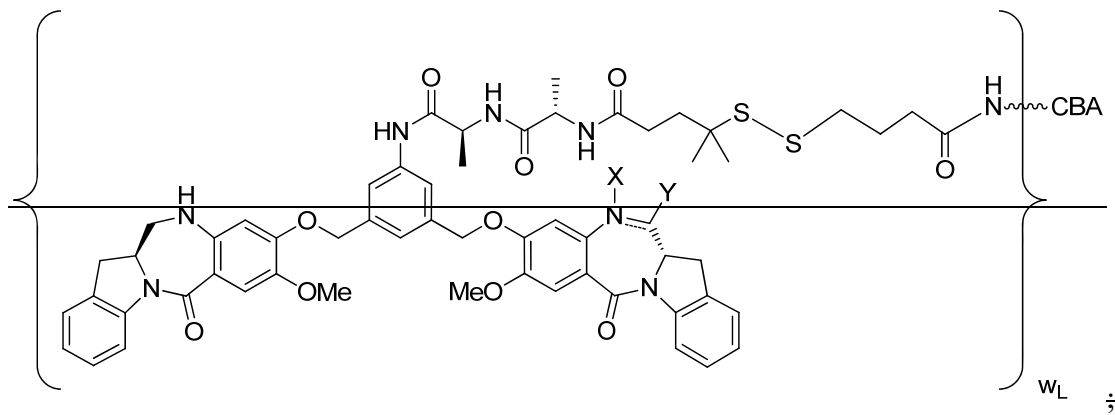
wherein:

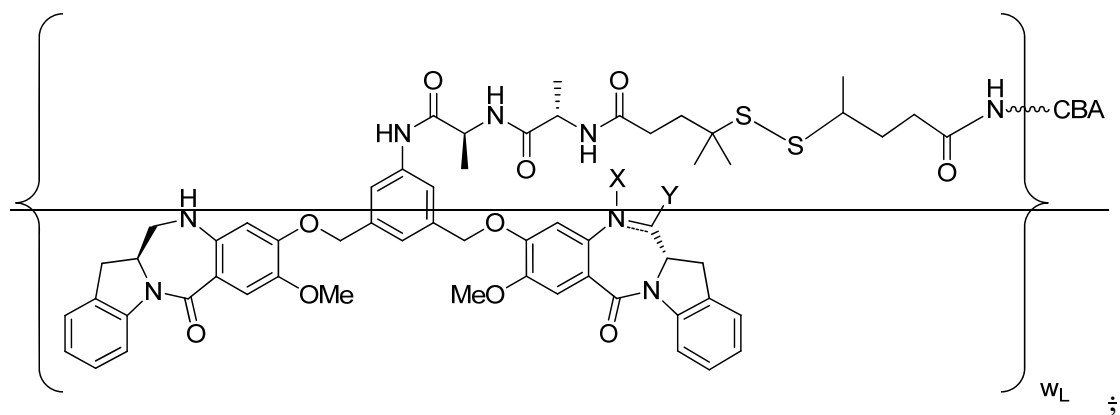
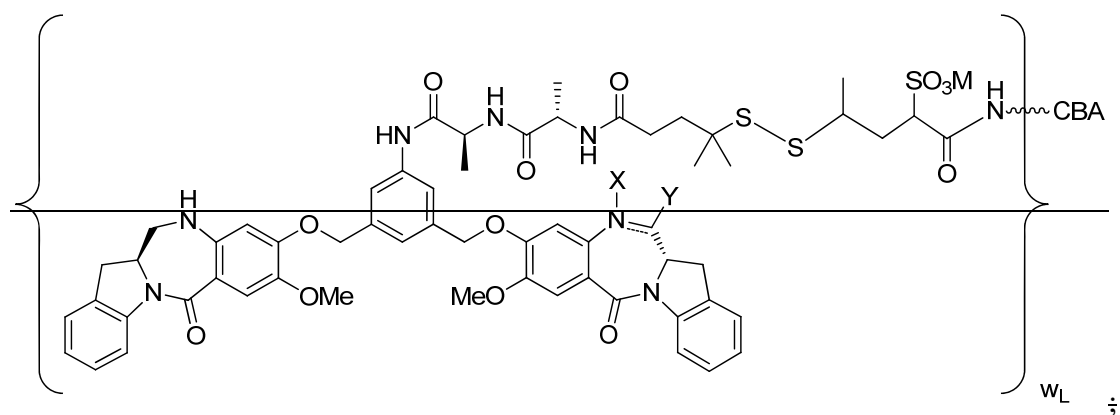
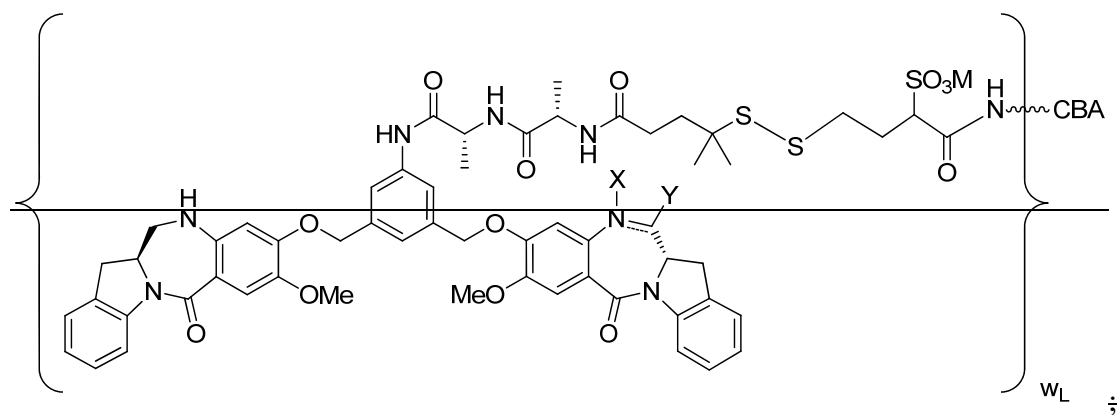
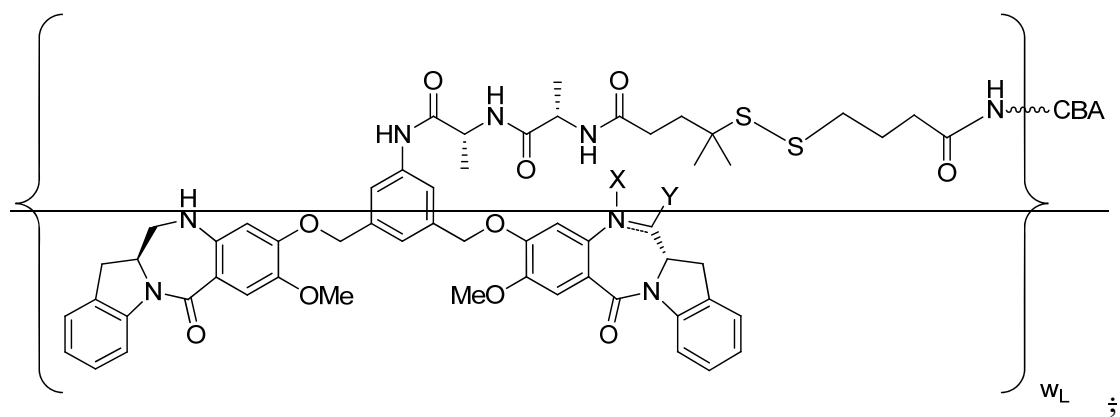
q is an integer from 1 to 5; and

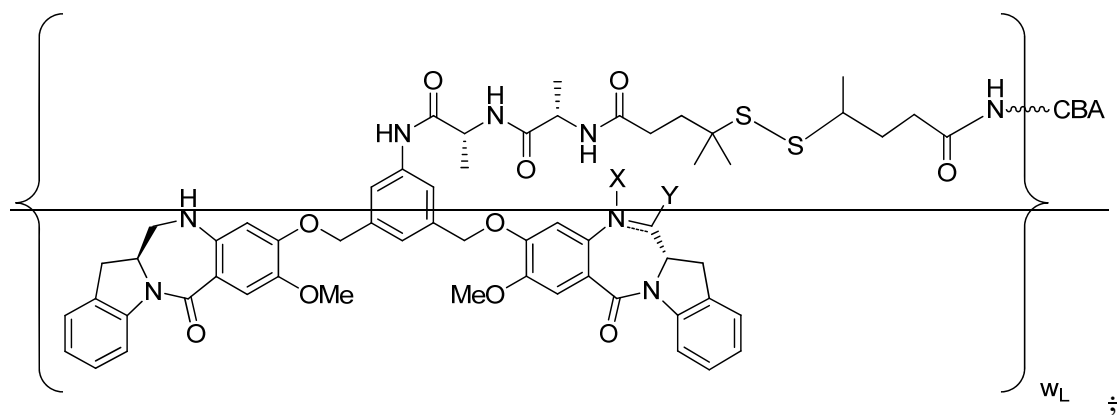
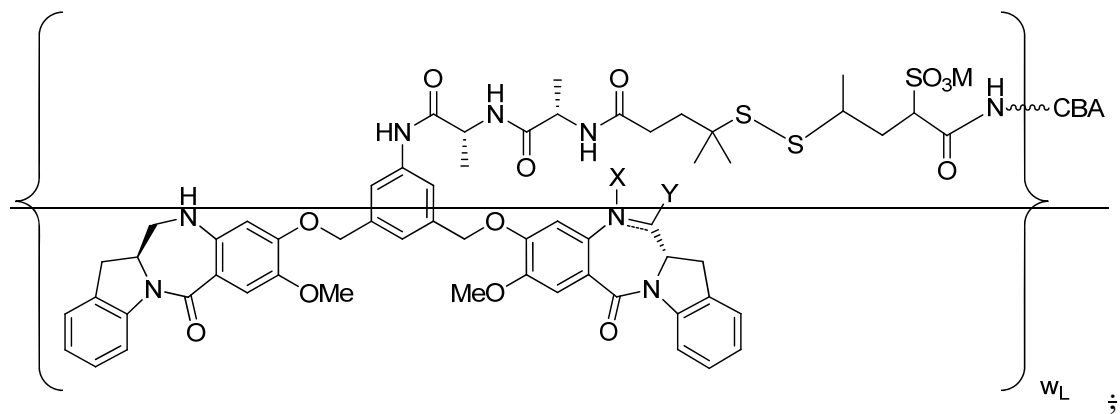
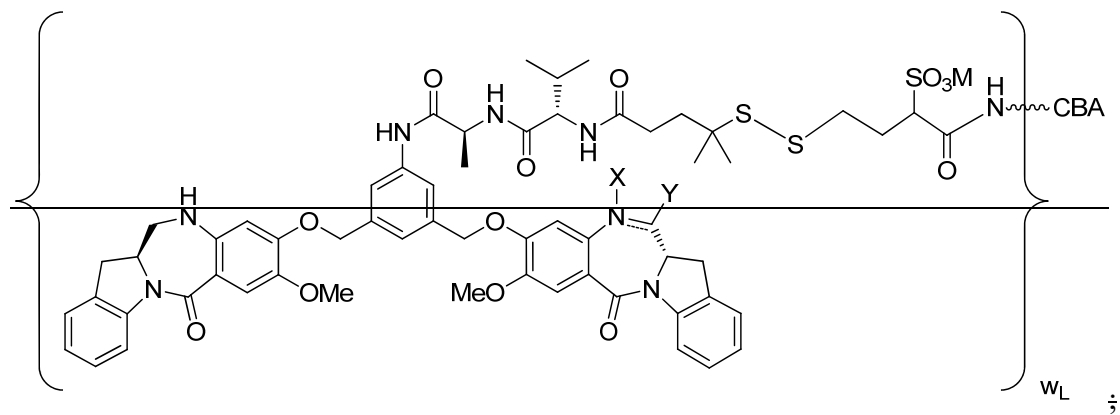
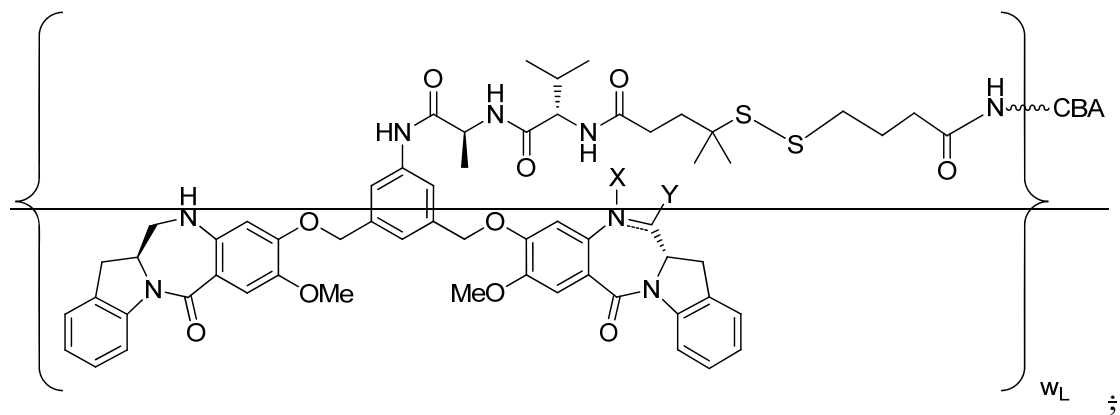
M is H<sup>+</sup> or a cation.

61. ~~The immunoconjugate of claim 60, wherein R<sub>a</sub> and R<sub>b</sub> are both H; and R<sub>s</sub> is H or Me.~~
62. ~~The immunoconjugate of claim 60 or 61, wherein P is a peptide containing 2 to 5 amino acid residues.~~
63. ~~The immunoconjugate of any one of claims 60-62, wherein P is selected from Gly-Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Ile-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl-Arg, Phe-N<sup>9</sup>-nitro-Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55), β-Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.~~
64. ~~The immunoconjugate of claim 63, wherein P is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, or D-Ala-D-Ala.~~
65. ~~The immunoconjugate of any one of claims 60-64, wherein Q is -SO<sub>3</sub>M.~~
66. ~~The immunoconjugate of claim 60, wherein the immunoconjugate is represented by the following formula:~~







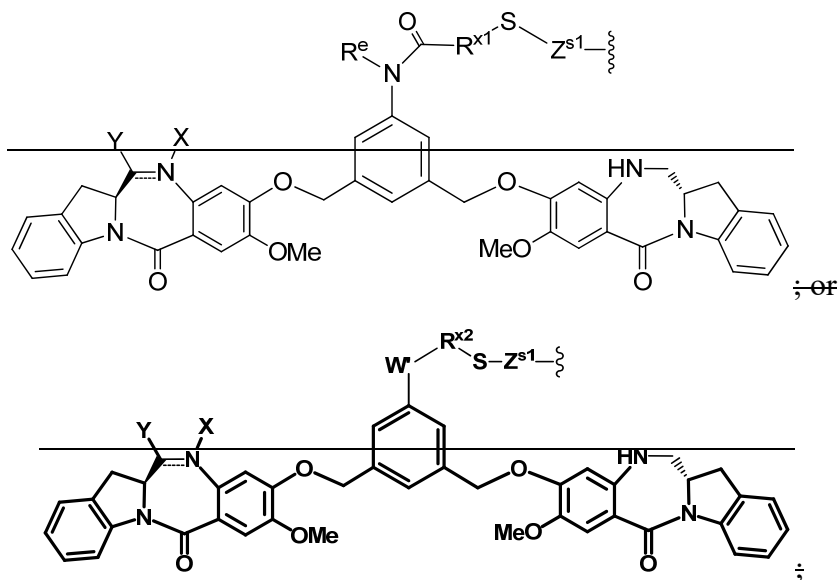




a lysine residue;

$W_L$  is an integer from 1 to 20; and

$Cy^{L2}$  is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line  $\equiv$  between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is H or an amine protecting moiety, and Y is OH or SO<sub>3</sub>M;

$R^{x1}$  and  $R^{x2}$  are independently (C<sub>1</sub>-C<sub>6</sub>)alkyl;

$R^e$  is H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

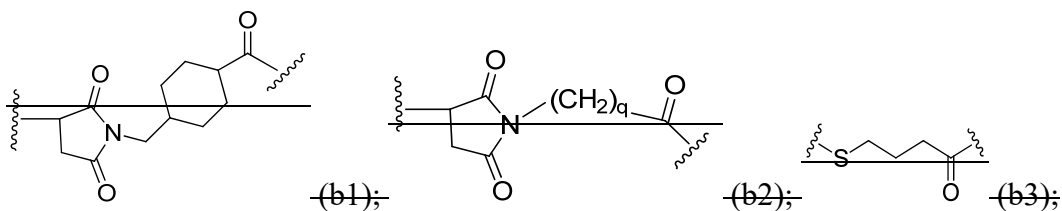
$W'$  is NR<sup>e2</sup>;

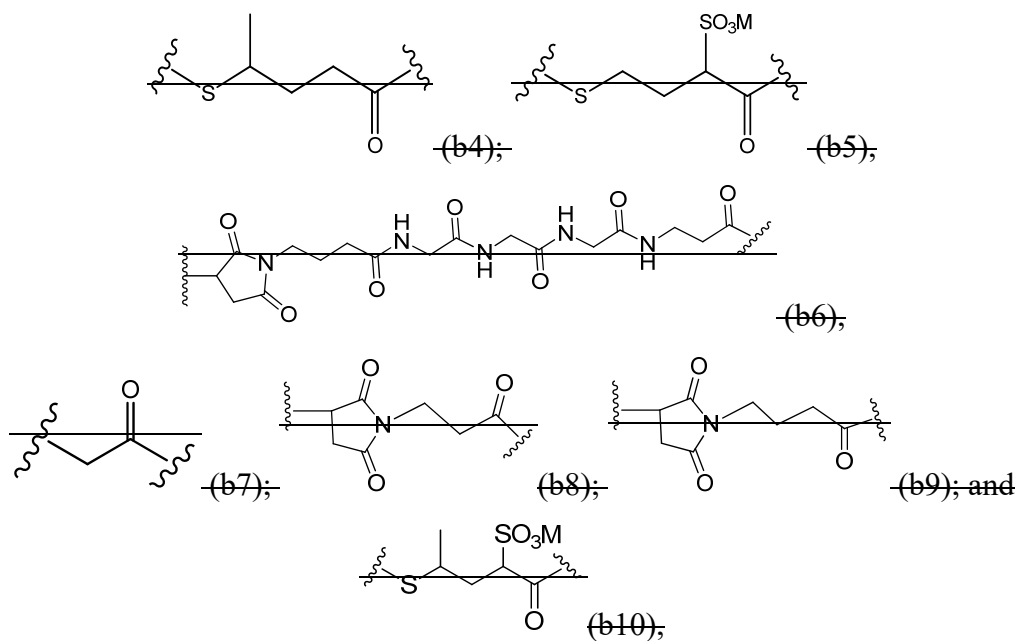
$R^{e2}$  is (CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is H or Me;

Z<sup>s1</sup> is selected from any one of the following formulas:



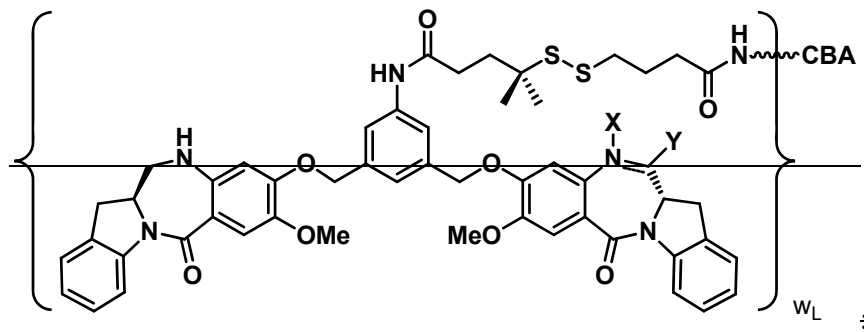


wherein:

q is an integer from 1 to 5; and

M is  $\text{H}^+$  or a cation.

68. The immunoconjugate of claim 67, wherein  $\text{R}^e$  is H or Me;  $\text{R}^{*1}$  and  $\text{R}^{*2}$  are independently  $(\text{CH}_2)_p(\text{CR}^f\text{R}^g)$ , wherein  $\text{R}^f$  and  $\text{R}^g$  are each independently H or a  $(\text{C}_1\text{-C}_4)$ alkyl; and p is 0, 1, 2 or 3.
69. The immunoconjugate of claim 68, wherein  $\text{R}^f$  and  $\text{R}^g$  are the same or different, and are selected from H and Me.
70. The immunoconjugate of claim 67, wherein the immunoconjugate is represented by the following formula:



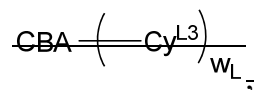


and C represents a double bond, X is absent and Y is H.

72. The immunoconjugate of any one of claims 60-70, wherein the double line  $\equiv$  between N and C represents a single bond, X is H, and Y is  $\text{SO}_3\text{M}$ .

73. The immunoconjugate of claim 72, wherein M is  $\text{H}^+$ ,  $\text{Na}^+$  or  $\text{K}^+$ .

74. An immunoconjugate having the formula:

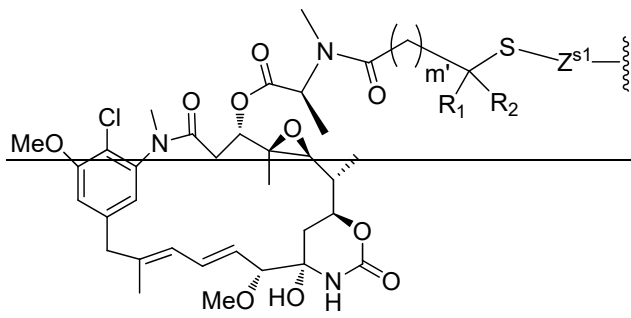


wherein:

CBA is an antibody or antigen binding fragment thereof of any one of claims 23-26, or the polypeptide of claim 55 or 56, which is covalently linked to  $\text{Cy}^{\text{L3}}$  through a Lys residue;

$\text{W}_\text{L}$  is an integer from 1 to 20;

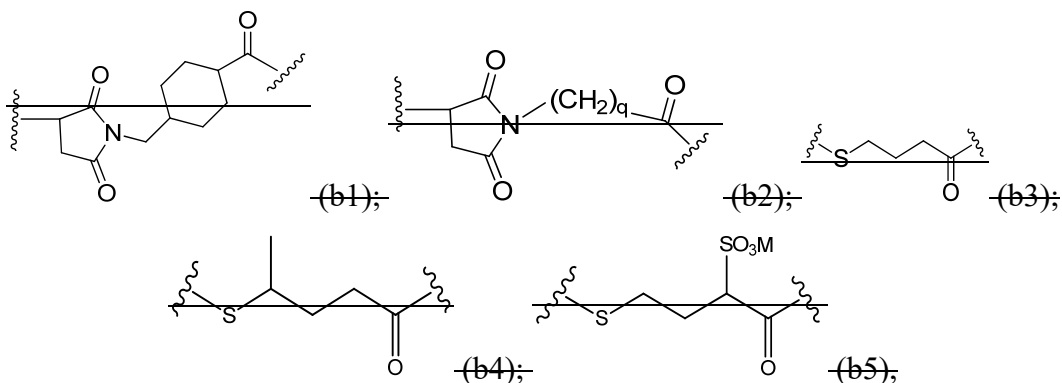
$\text{Cy}^{\text{L3}}$  is represented by the following formula:

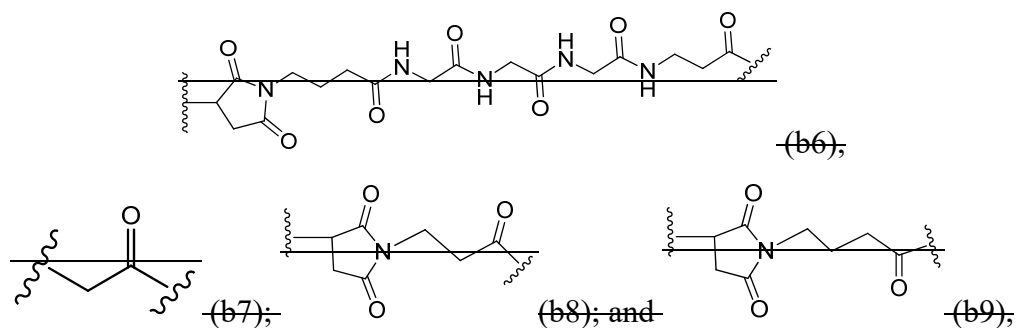


$m'$  is 1 or 2;

$\text{R}_1$  and  $\text{R}_2$  are each independently H or a  $(\text{C}_1\text{-C}_3)$  alkyl; and

$\text{Z}^{\text{s1}}$  is selected from any one of the following formulas:





wherein:

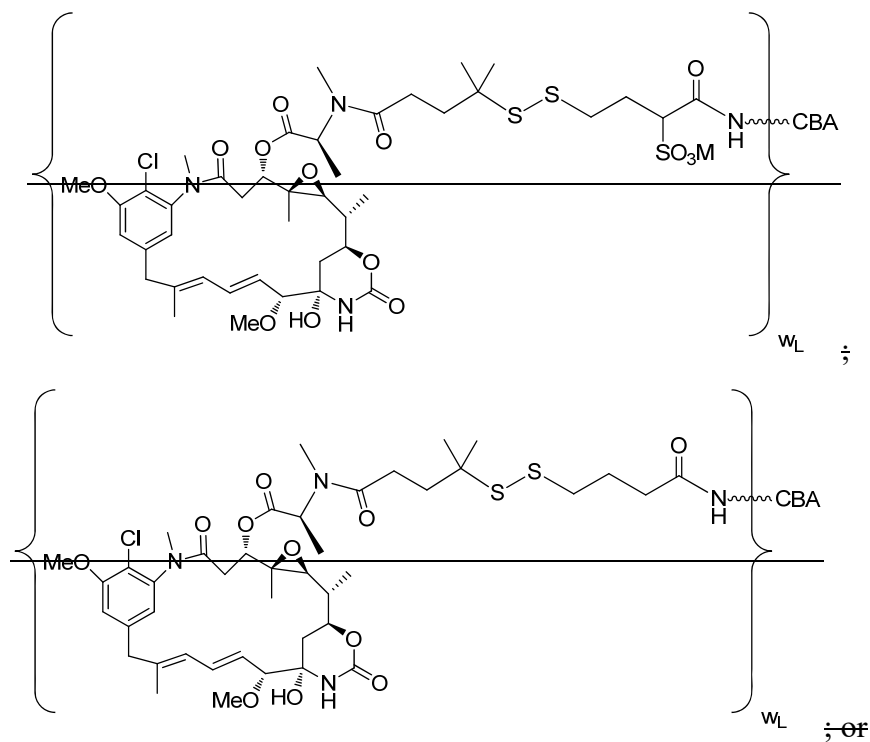
q is an integer from 1 to 5; and

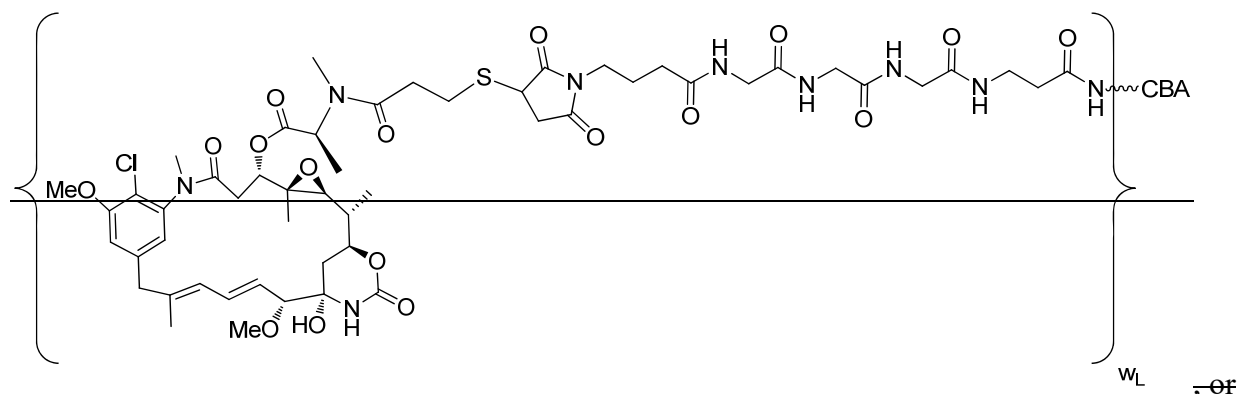
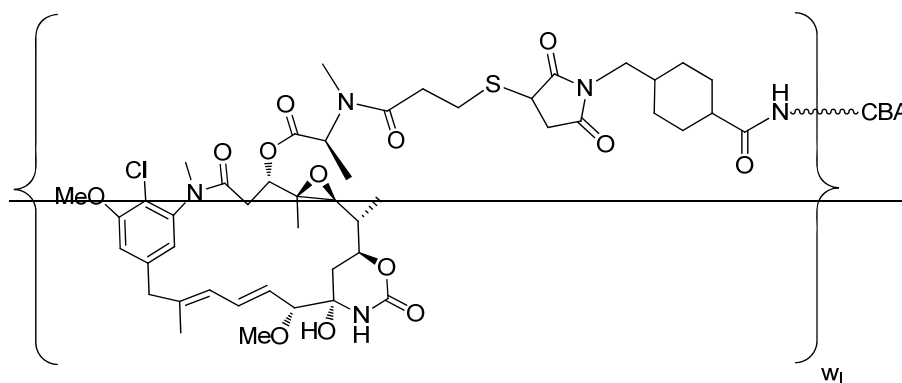
M is  $H^+$  or a cation.

75. ~~The immunoconjugate of claim 74, wherein  $m'$  is 1, and  $R_1$  and  $R_2$  are both H.~~

76. ~~The immunoconjugate of claim 74, wherein  $m'$  is 2, and  $R_1$  and  $R_2$  are both Me.~~

77. ~~The immunoconjugate of claim 74, wherein the immunoconjugate is represented by the following formula:~~

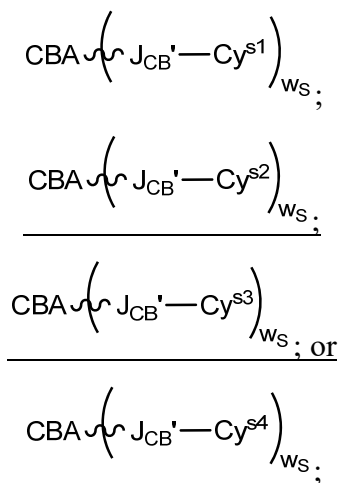




or a pharmaceutically acceptable salt thereof, wherein  $W_L$  is an integer from 1 to 10.

78. The immunoconjugate of claim 77, wherein M is  $H^+$ ,  $Na^+$  or  $K^+$ .

17 79. An immunoconjugate having the following formula:



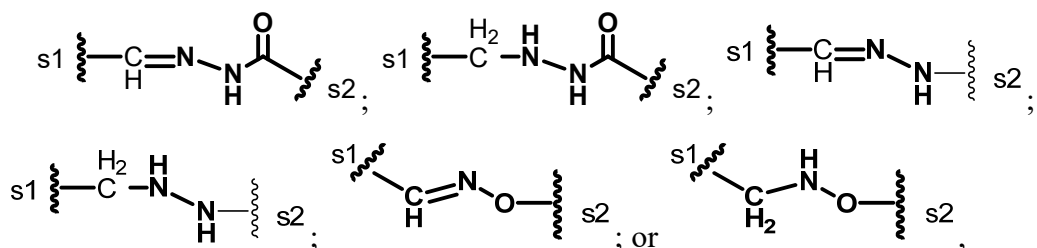
wherein:

CBA is an the antibody or antigen-binding fragment thereof of claim 7 any one of claims 27-36, or the polypeptide thereof of claim 13 55 or 56, covalently linked to the

JCB' group;

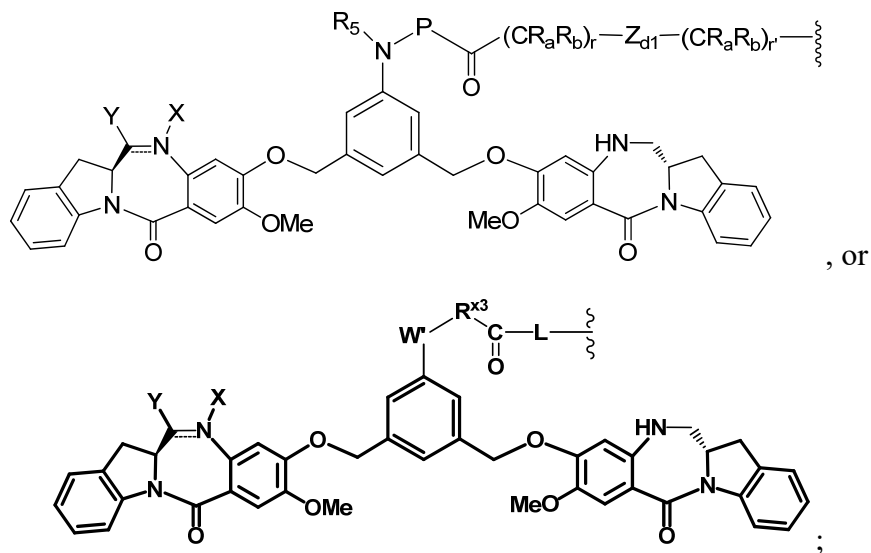
$W_S$  is 1, 2, 3, or 4;

J<sub>CB'</sub> is a moiety formed by reacting an aldehyde group derived from oxidation of a 2-hydroxyethylamine moiety on an N-terminal of said antibody or antigen-binding fragment thereof ~~of any one of claims 27-36, or the said polypeptide thereof of claim 55 or 56,~~ and an aldehyde reactive group on Cy<sup>s1</sup>, Cy<sup>s2</sup>, Cy<sup>s3</sup>, or Cy<sup>s4</sup> and is represented by the following formula:



wherein s1 is the site covalently linked to the CBA; and s2 is the site covalently linked to Cy<sup>s1</sup>;

Cy<sup>s1</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line == between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is -H or an amine protecting moiety, Y is -OH or -SO<sub>3</sub>M, and M is H<sup>+</sup> or a cation;

R<sub>5</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

P is an amino acid residue or a peptide containing 2 to 20 amino acid residues;

Z<sub>d1</sub> is absent, -C(=O)-NR<sub>9</sub>-, or -NR<sub>9</sub>-C(=O)-;

R<sub>9</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

R<sub>a</sub> and R<sub>b</sub>, for each occurrence, are independently -H, (C<sub>1</sub>-C<sub>3</sub>)alkyl, or a charged substituent or an ionizable group Q;

r and r' are independently an integer from 1 to 6;

W' is -NR<sup>e'</sup>,

R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

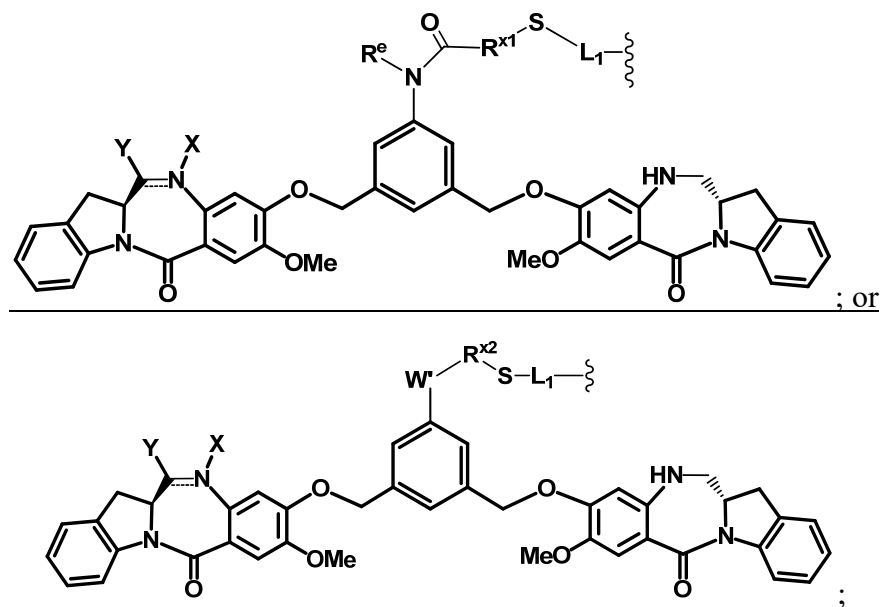
R<sup>k</sup> is -H or -Me;

R<sup>x3</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

L is -NR<sub>9</sub>-(CR<sub>a</sub>R<sub>b</sub>)<sub>r''</sub> or absent; and

r'' is an integer from 0 to 6;

Cy<sup>s2</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line == between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is -H or an amine protecting moiety, and Y is -OH or -SO<sub>3</sub>M;

M is H<sup>+</sup> or a cation;

R<sup>x1</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

R<sup>e</sup> is -H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

W' is -NR<sup>e'</sup>,

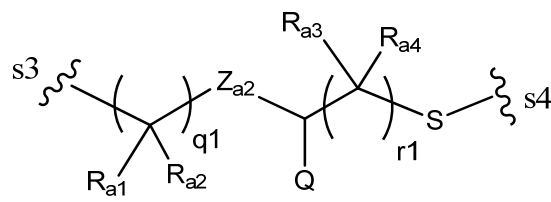
R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is -H or -Me;

R<sup>x2</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

L<sub>1</sub> is represented by the following formula:



wherein:

s<sub>3</sub> is the site covalently linked to the group J<sub>CB</sub>';

s<sub>4</sub> is the site covalently linked to the -S- group on Cy<sup>s2</sup>;

Z<sub>a2</sub> is absent, -C(=O)-NR<sub>9</sub>-, or -NR<sub>9</sub>-C(=O)-;

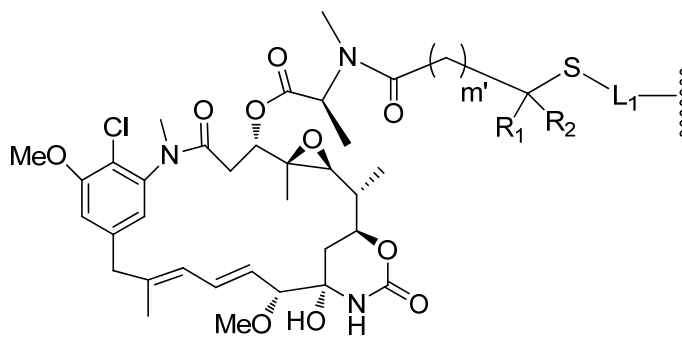
R<sub>9</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Q is H, a charged substituent or an ionizable group;

R<sub>a1</sub>, R<sub>a2</sub>, R<sub>a3</sub>, R<sub>a4</sub>, for each occurrence, are independently H or (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

q<sub>1</sub> and r<sub>1</sub> are each independently an integer from 0 to 10, provided that q<sub>1</sub> and r<sub>1</sub> are not both 0;

Cy<sup>s3</sup> is represented by the following formula:

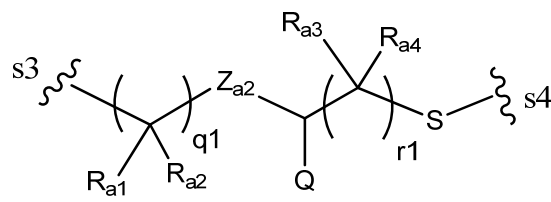


wherein:

m' is 1 or 2;

R<sub>1</sub> and R<sub>2</sub>, are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

L<sub>1</sub> is represented by the following formula:



wherein:

s<sub>3</sub> is the site covalently linked to the group J<sub>CB</sub>';

s<sub>4</sub> is the site covalently linked to the -S- group on Cy<sup>s<sub>3</sub></sup>;

Z<sub>a2</sub> is absent, -C(=O)-NR<sub>9</sub>-, or -NR<sub>9</sub>-C(=O)-;

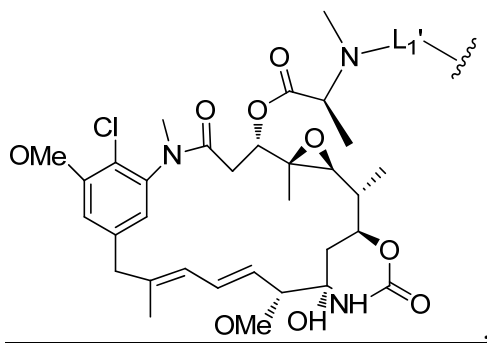
R<sub>9</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Q is H, a charged substituent or an ionizable group;

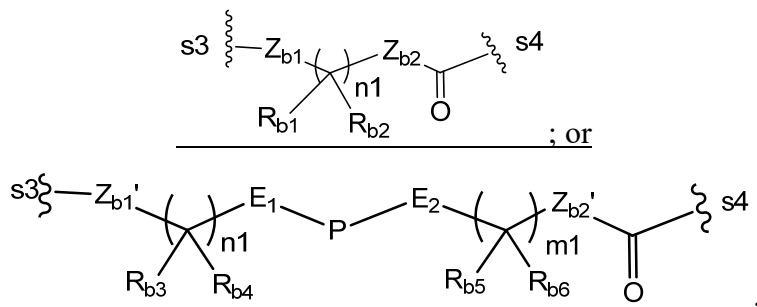
R<sub>a1</sub>, R<sub>a2</sub>, R<sub>a3</sub>, R<sub>a4</sub>, for each occurrence, are independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

q<sub>1</sub> and r<sub>1</sub> are each independently an integer from 0 to 10, provided that q<sub>1</sub> and r<sub>1</sub> are not both 0;

Cy<sup>s<sub>4</sub></sup> is represented by the following formula:



L<sub>1</sub>' is represented by the following formula:



wherein:

s<sub>3</sub> is the site covalently linked to the group J<sub>CB</sub>' group;

s<sub>4</sub> is the site covalently linked to -NMe- group on Cy<sup>s<sub>4</sub></sup>;

Z<sub>b1</sub> and Z<sub>b2</sub> are both absent, or one of Z<sub>b1</sub> and Z<sub>b2</sub> is absent and the other is -CH<sub>2</sub>-O- or -O-CH<sub>2</sub>-;

Z<sub>b1</sub>' and Z<sub>b2</sub>' are each independently absent, -CH<sub>2</sub>-O-, -O-CH<sub>2</sub>-, -NR<sub>9</sub>-C(=O)-CH<sub>2</sub>-, or -CH<sub>2</sub>-C(=O)-NR<sub>9</sub>-;

R<sub>9</sub> is H or (C<sub>1</sub>-C<sub>3</sub>)alkyl;

n<sub>1</sub> and m<sub>1</sub> are each independently an integer from 1 to 6;

one of E<sub>1</sub> and E<sub>2</sub> is -C(=O)-, and the other is -NR<sub>9</sub>-; or one of E<sub>1</sub> and E<sub>2</sub> is -C(=O)- or -NR<sub>9</sub>-, and the other is absent;

P is an amino acid residue or a peptide containing between 2 to 20 amino acid residues; and

R<sub>b1</sub>, R<sub>b2</sub>, R<sub>b3</sub>, R<sub>b4</sub>, R<sub>b5</sub> and R<sub>b6</sub>, for each occurrence, are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl.

~~80. The immunoconjugate of claim 79, wherein R<sub>a</sub> and R<sub>b</sub> are both H, and R<sub>s</sub> and R<sub>9</sub> are both H or Me.~~

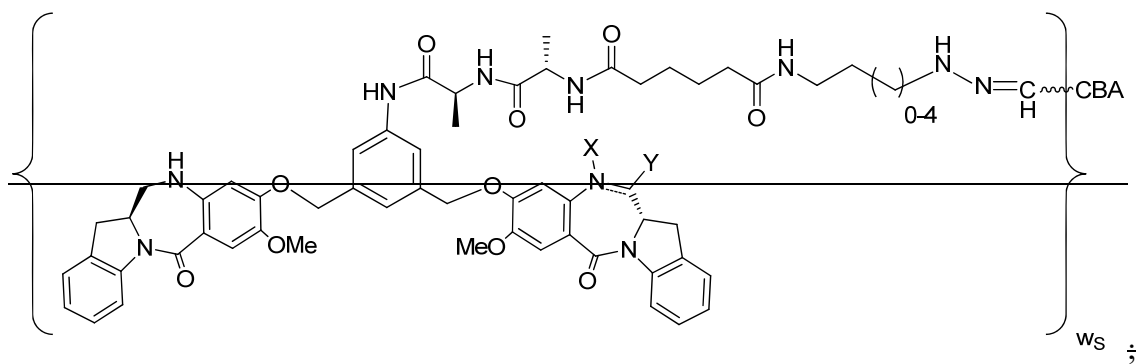
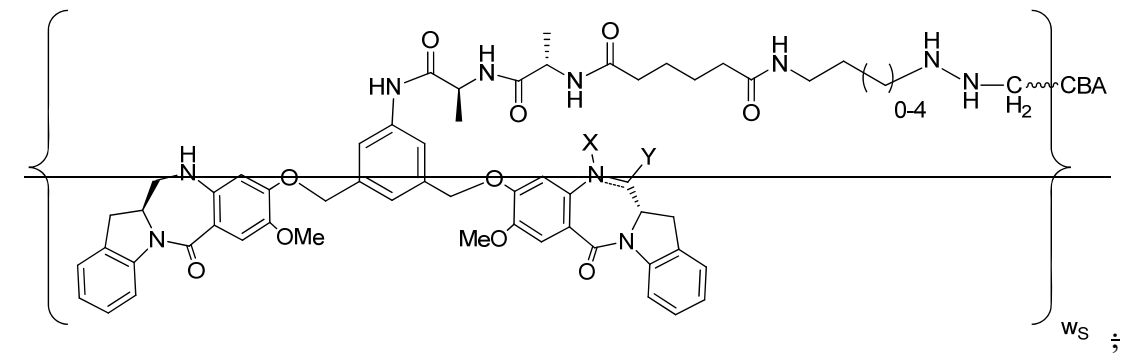
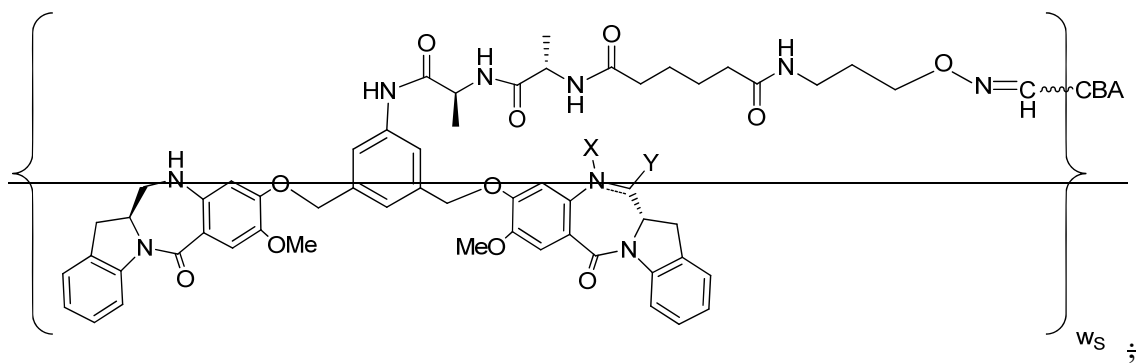
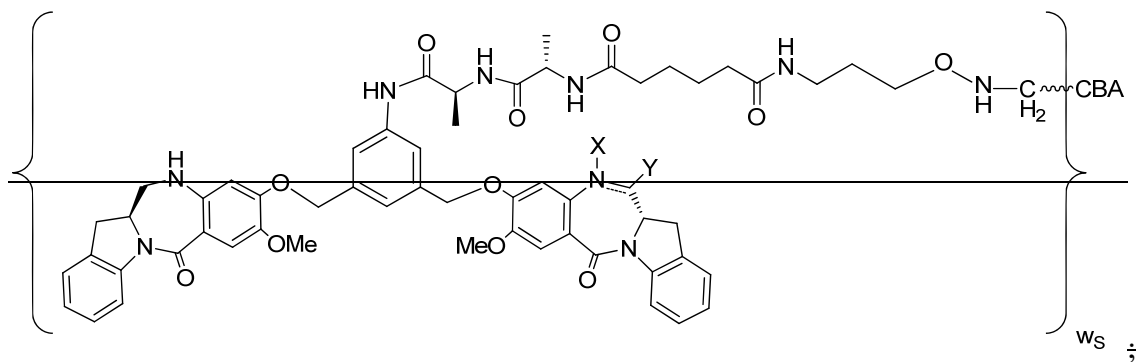
~~81. The immunoconjugate of claim 79 or 80, wherein P is a peptide containing 2 to 5 amino acid residues.~~

~~82. The immunoconjugate of any one of claims 79-81, wherein P is selected from the group consisting of: Gly-Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Ile-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl-Arg, Phe-N<sup>9</sup>-nitro-Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55), β-Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.~~

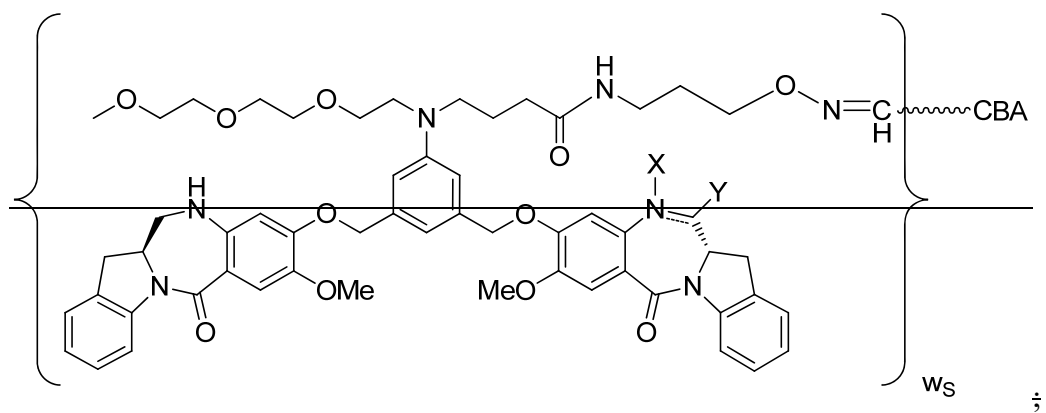
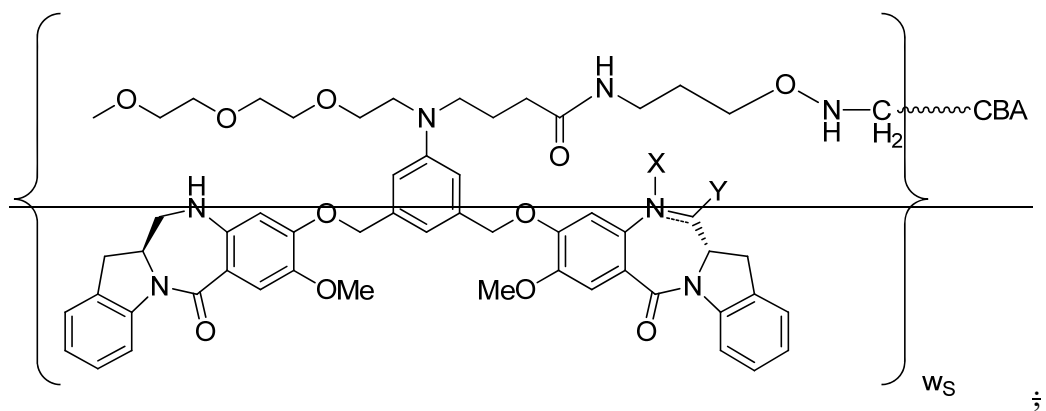
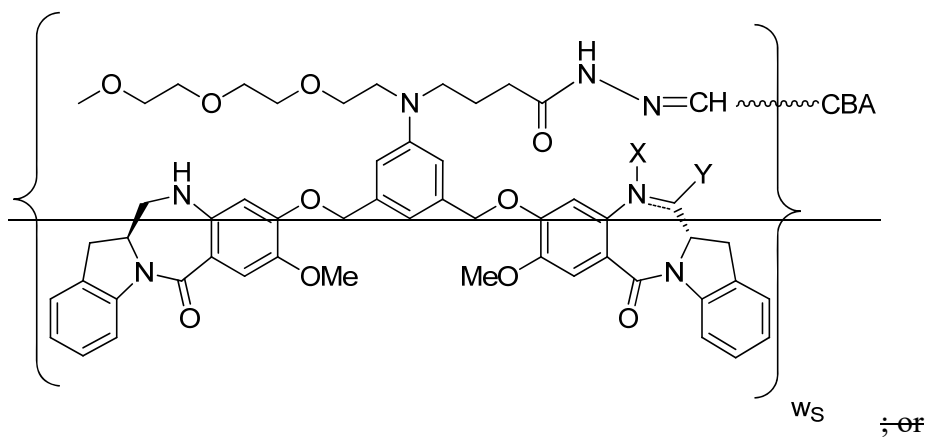
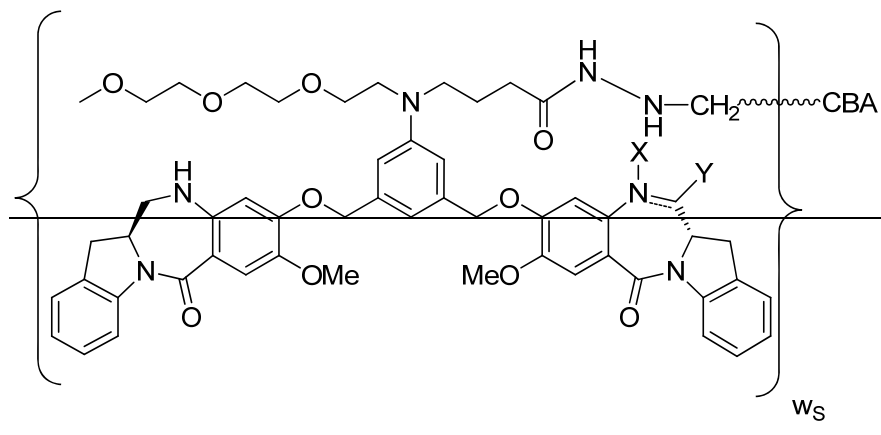
~~83. The immunoconjugate of claim 82, wherein P is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, or D-Ala-D-Ala.~~

~~84. The immunoconjugate of any one of claims 79-83, wherein Q is -SO<sub>3</sub>M.~~

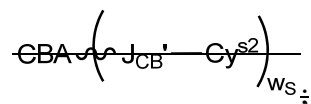
~~85. The immunoconjugate of claim 80, which is represented by the following formula:~~





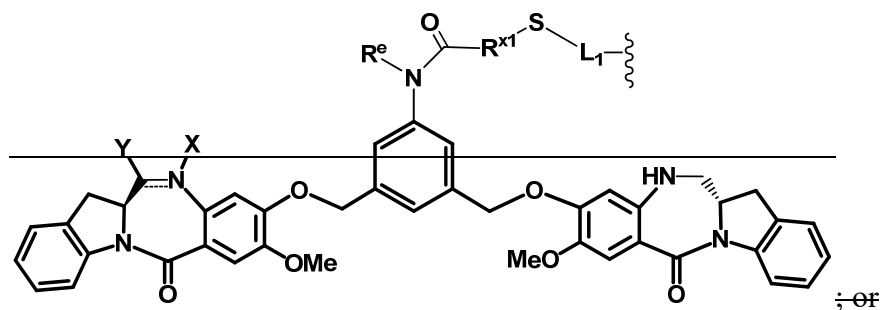


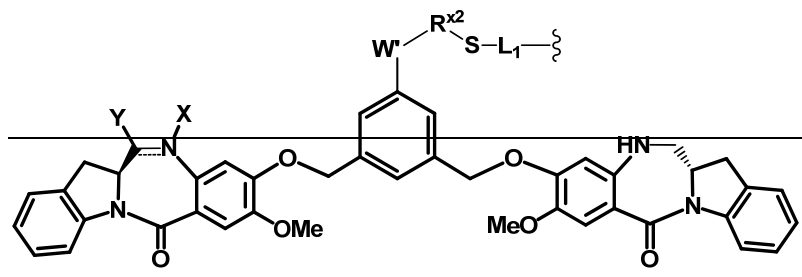
~~86. An immunoconjugate having the following formula:~~



~~CBA is the antibody or antigen-binding fragment thereof of any one of claims 27-36, or the polypeptide thereof of any claim 55 or 56, covalently linked to the JCB' group,~~

~~Cy<sup>s2</sup> is represented by the following formula:~~





or a pharmaceutically acceptable salt thereof, wherein:

the double line  $\equiv$  between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is H or an amine protecting moiety, and Y is OH or SO<sub>3</sub>M;

M is H<sup>+</sup> or a cation;

R<sup>x1</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

R<sup>e</sup> is H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

W' is NR<sup>e2</sup>;

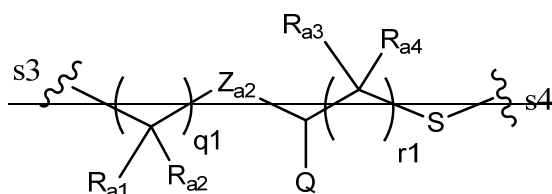
R<sup>e2</sup> is (CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is H or Me;

R<sup>x2</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

L<sub>1</sub> is represented by the following formula:



wherein:

s3 is the site covalently linked to the group J<sub>CB</sub>';

s4 is the site covalently linked to the S group on Cy<sup>s2</sup>;

Z<sub>a2</sub> is absent, C(=O)NR<sub>9</sub>, or NR<sub>9</sub>C(=O);

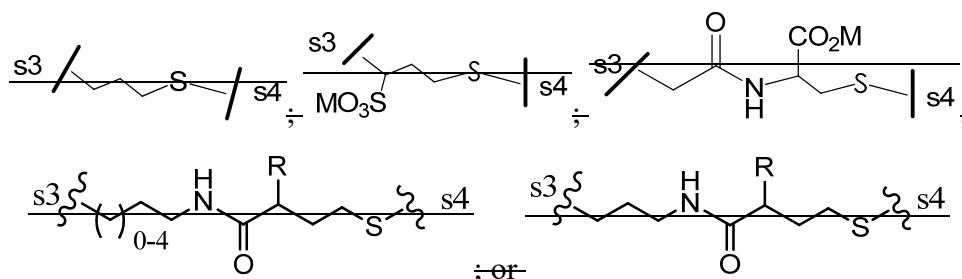
R<sub>9</sub> is H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Q is H, a charged substituent or an ionizable group;

R<sub>a1</sub>, R<sub>a2</sub>, R<sub>a3</sub>, R<sub>a4</sub>, for each occurrence, are independently H or (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

q1 and r1 are each independently an integer from 0 to 10, provided that q1 and r1 are not both 0.

87. The immunoconjugate of claim 86, wherein  $L_1$  is represented by the following formula:

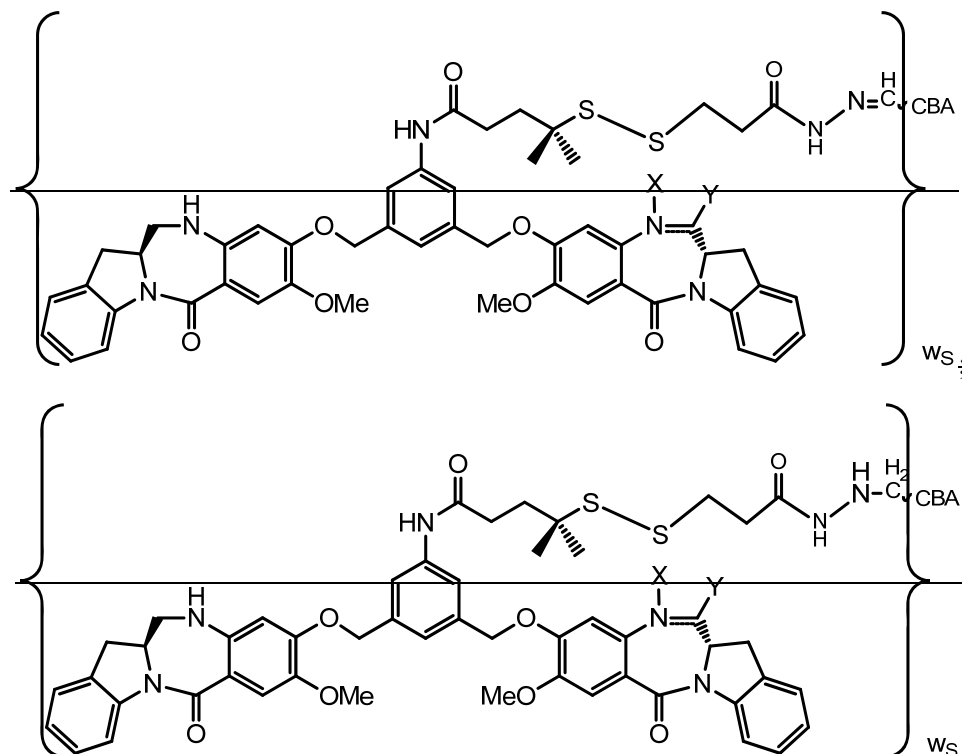


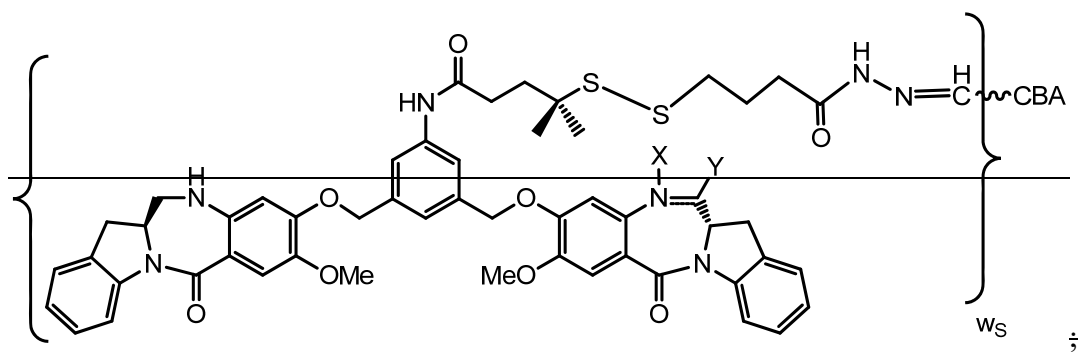
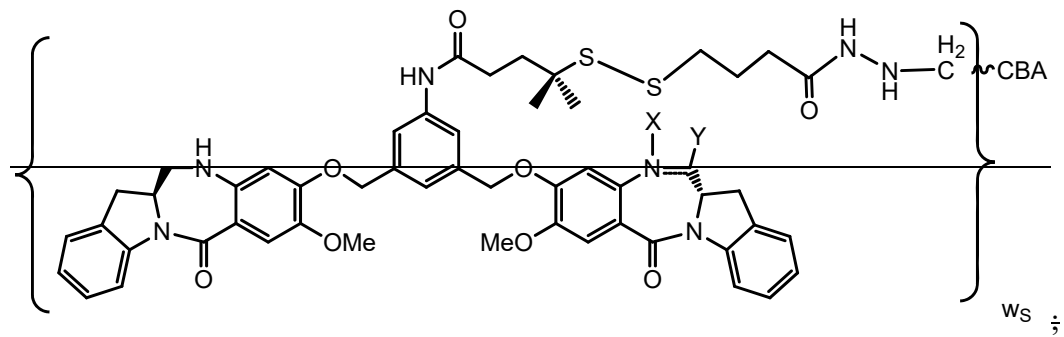
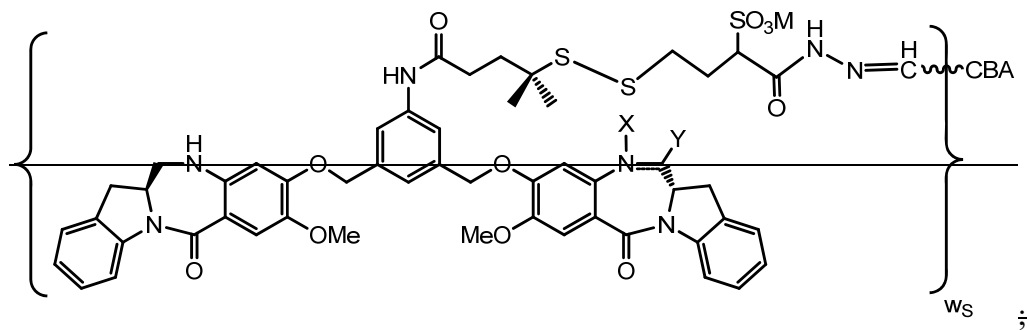
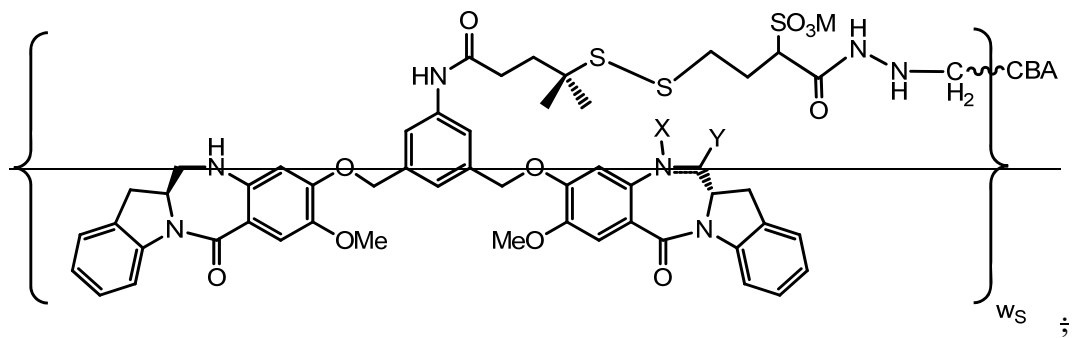
or a pharmaceutically acceptable salt thereof, wherein R is H or  $\text{SO}_3\text{M}$ .

88. The immunoconjugate of claim 86 or 87, wherein  $R^*$  is H or Me; and  $R^{*1}$  is  $(\text{CH}_2)_p$ - $(\text{CR}^f\text{R}^g)$ , and  $R^{*2}$  is  $(\text{CH}_2)_p$ - $(\text{CR}^f\text{R}^g)$ , wherein  $R^f$  and  $R^g$  are each independently H or a  $(\text{C}_1\text{-C}_4)$ alkyl; and p is 0, 1, 2 or 3.

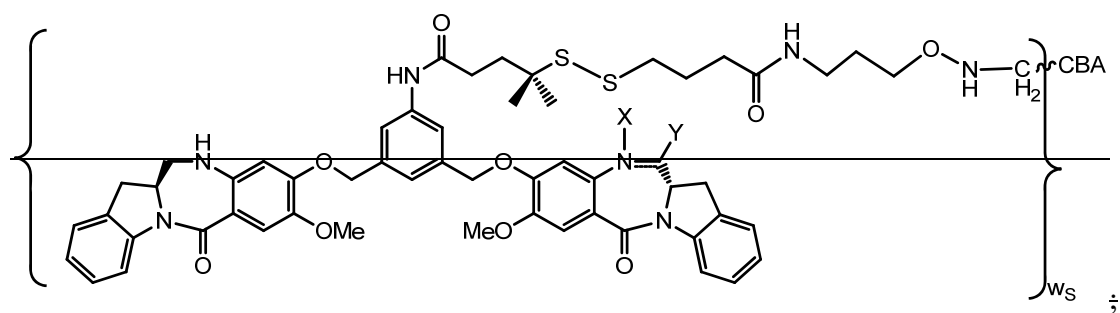
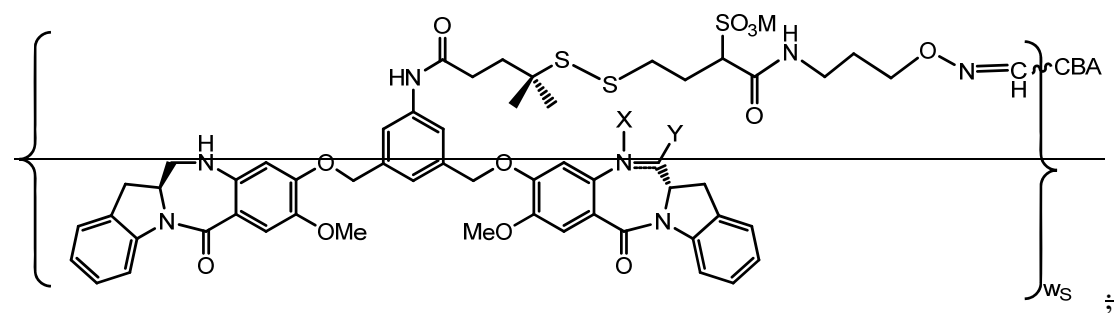
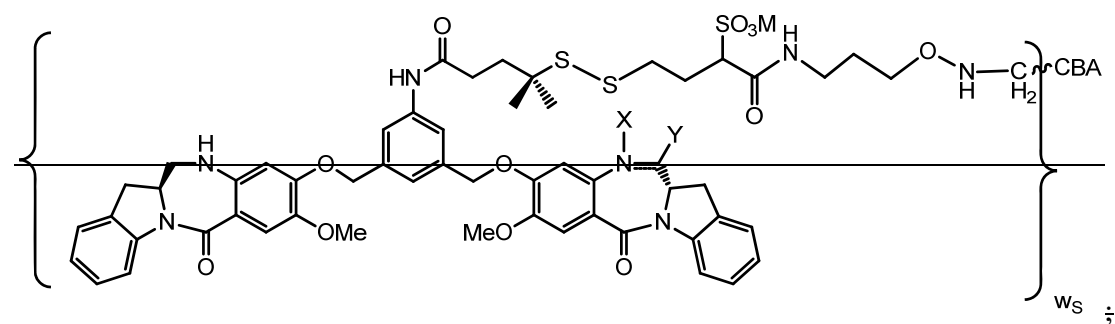
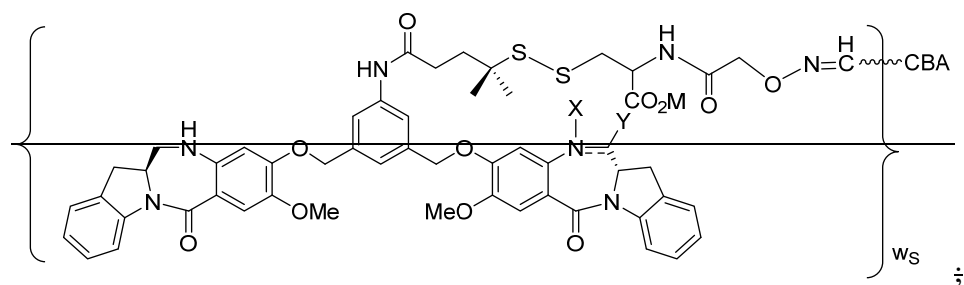
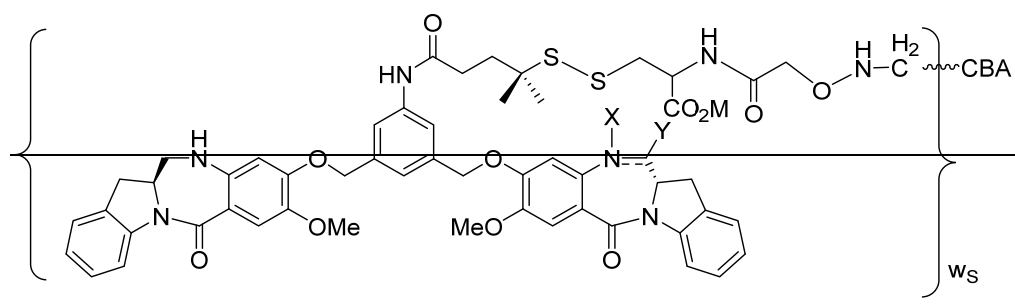
89. The immunoconjugate of claim 88, wherein  $R^f$  and  $R^g$  are the same or different, and are selected from H and Me.

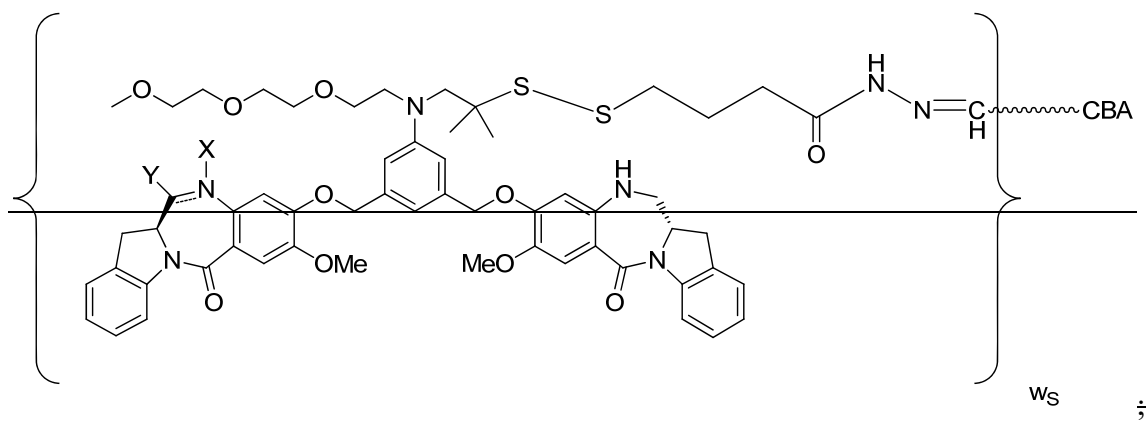
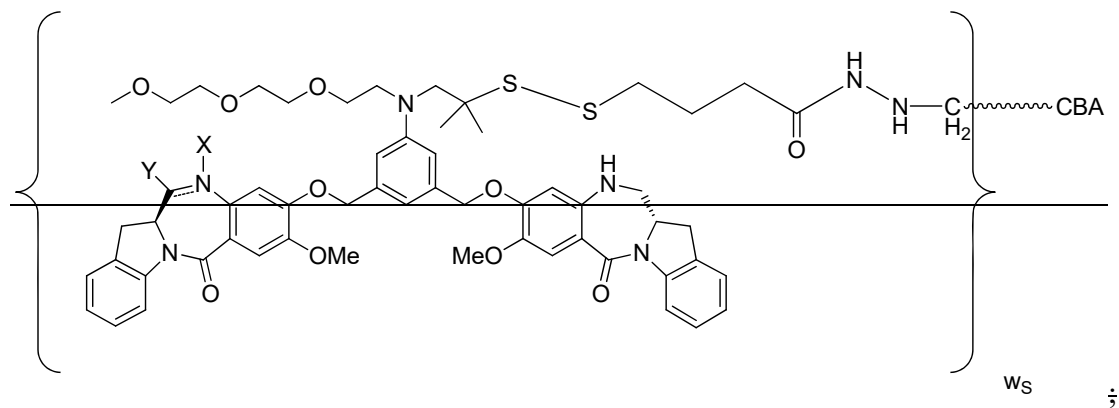
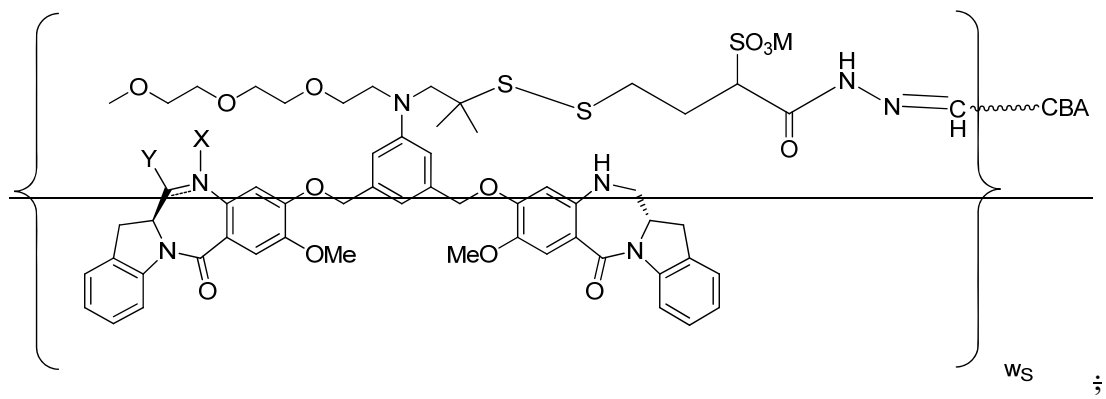
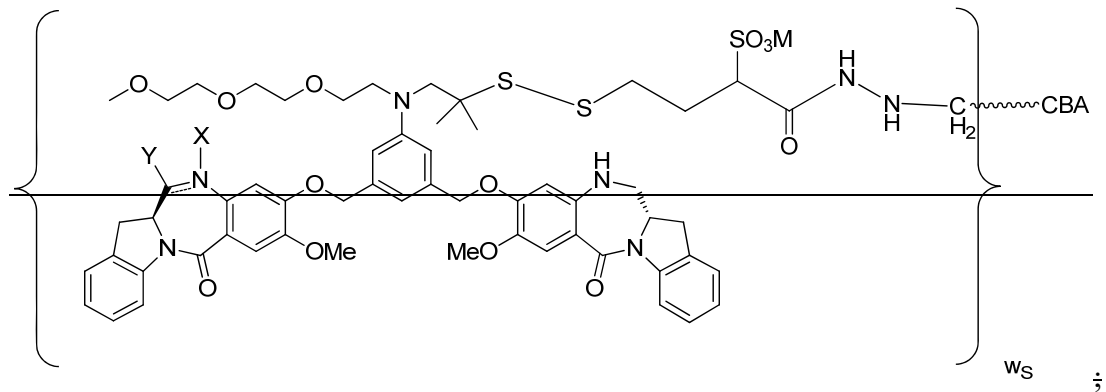
90. The immunoconjugate of claim 86, which is represented by the following formula:

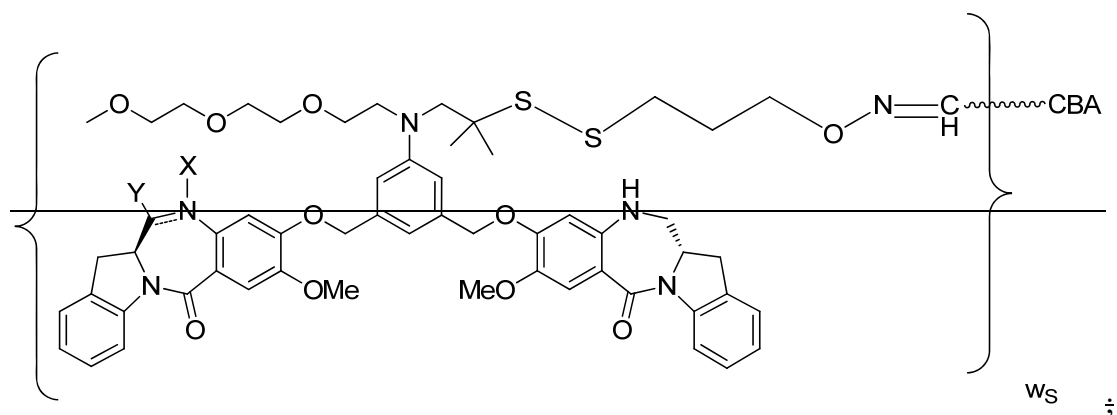
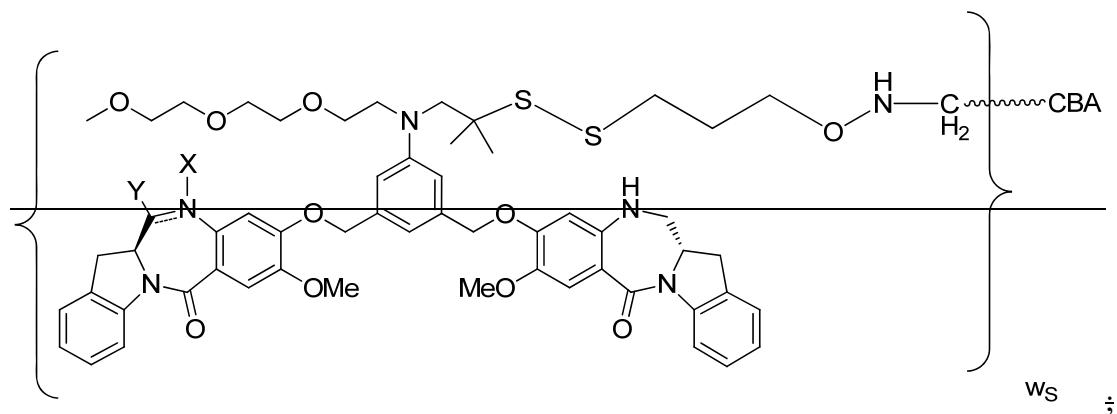
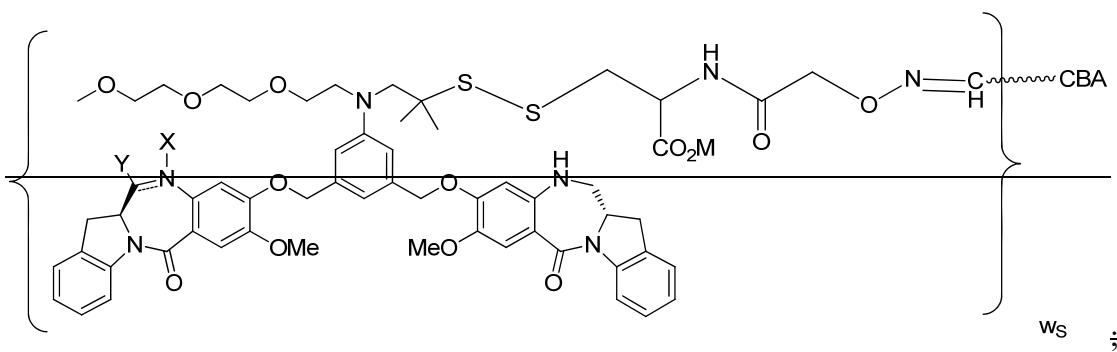
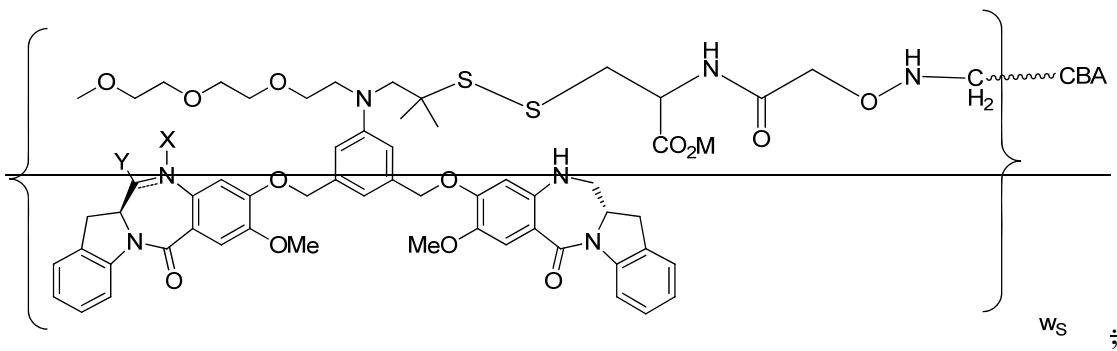


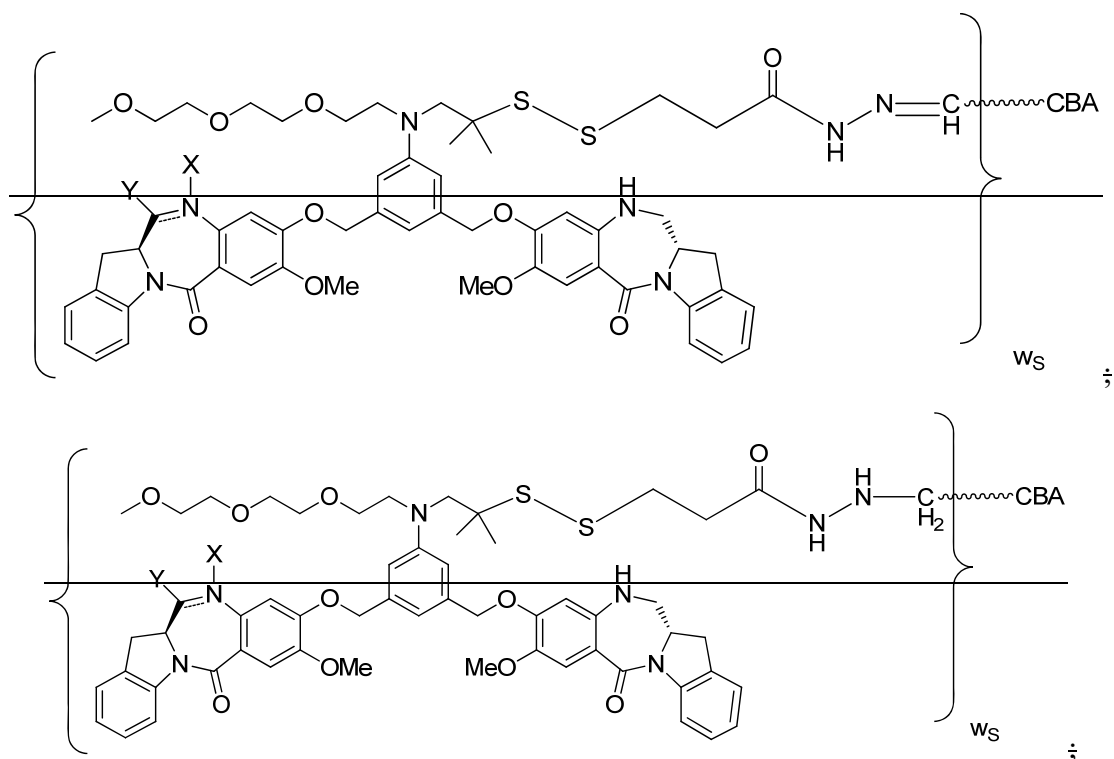






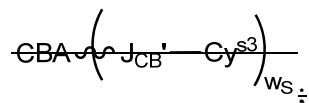






or a pharmaceutically acceptable salt thereof, wherein the double line  $\equiv$  between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is H; and when it is a single bond, X is H; and Y is OH or  $SO_3M$ .

91. The immunoconjugate of any one of claims 70-90, wherein the double line  $\equiv$  between N and C represents a double bond, X is absent and Y is H.
92. The immunoconjugate of any one of claims 79-90, wherein the double line  $\equiv$  between N and C represents a single bond, X is H and Y is  $SO_3M$ .
93. The immunoconjugate of claim 92, wherein M is  $H^+$ ,  $Na^+$  or  $K^+$ .
94. An immunoconjugate having the following formula:

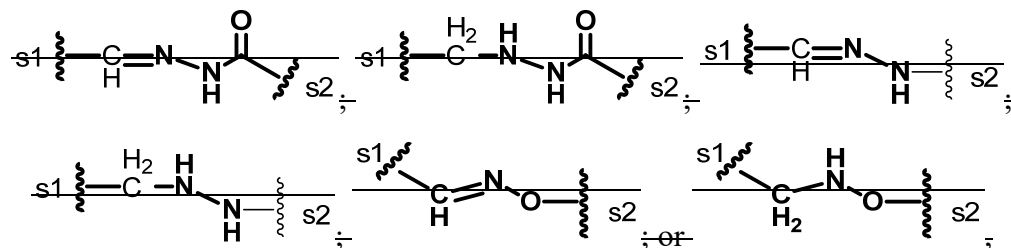


wherein:

CBA is the antibody or antigen binding fragment thereof of any one of claims 27-36, or the polypeptide thereof of claim 55 or 56, covalently linked to the  $J_{CB'}$  group;

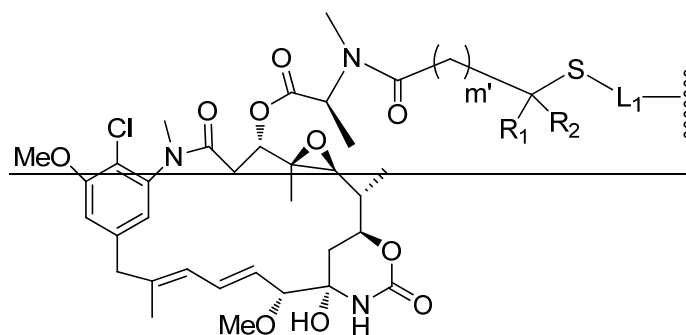
$J_{CB'}$  is a moiety formed by reacting an aldehyde group derived from oxidation of a 2-hydroxyethylamine moiety on an N-terminal of said antibody or antigen binding

fragment thereof of any one of claims 27-36, or the polypeptide thereof of claim 55 or 56, and an aldehyde reactive group on Cy<sup>s3</sup>, and is represented by the following formula:



wherein s1 is the site covalently linked to the CBA; and s2 is the site covalently linked to Cy<sup>s3</sup>;

Cy<sup>s3</sup> is represented by the following formula:

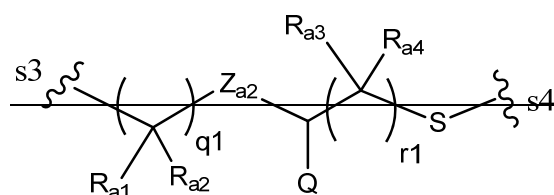


wherein:

m' is 1 or 2;

R<sub>1</sub> and R<sub>2</sub> are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

L<sub>1</sub> is represented by the following formula:



wherein:

s3 is the site covalently linked to the group J<sub>CB</sub>';

s4 is the site covalently linked to the S group on Cy<sup>s3</sup>;

Z<sub>a2</sub> is absent, C(=O)NR<sub>0</sub>, or NR<sub>0</sub>C(=O);

R<sub>0</sub> is H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Q is H, a charged substituent or an ionizable group;

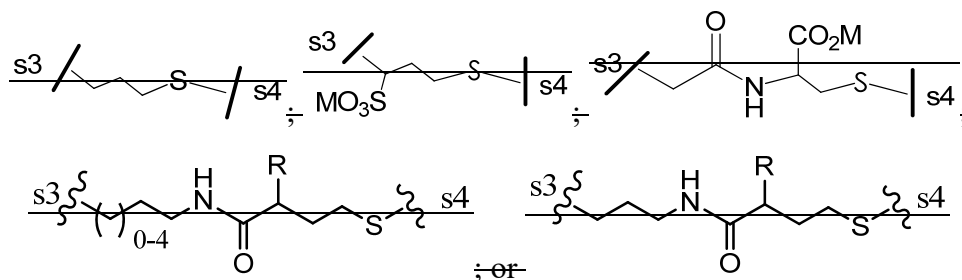
R<sub>a1</sub>, R<sub>a2</sub>, R<sub>a3</sub>, R<sub>a4</sub>, for each occurrence, are independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

q1 and r1 are each independently an integer from 0 to 10, provided that q1 and r1 are not both 0.

95. The immunoconjugate of claim 94, wherein m' is 1 and R<sub>1</sub> and R<sub>2</sub> are both H.

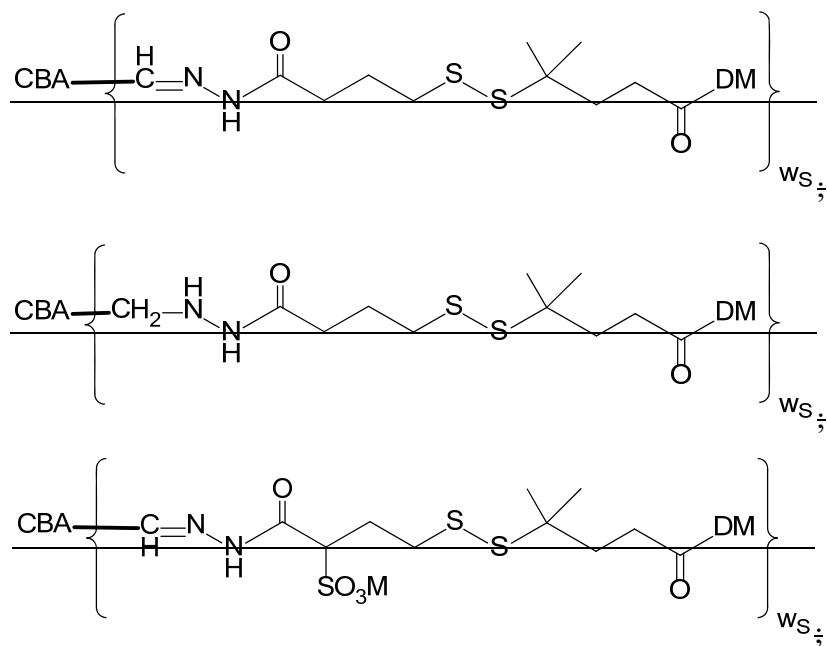
96. The immunoconjugate of claim 94, wherein m' is 2 and R<sub>1</sub> and R<sub>2</sub> are both Me.

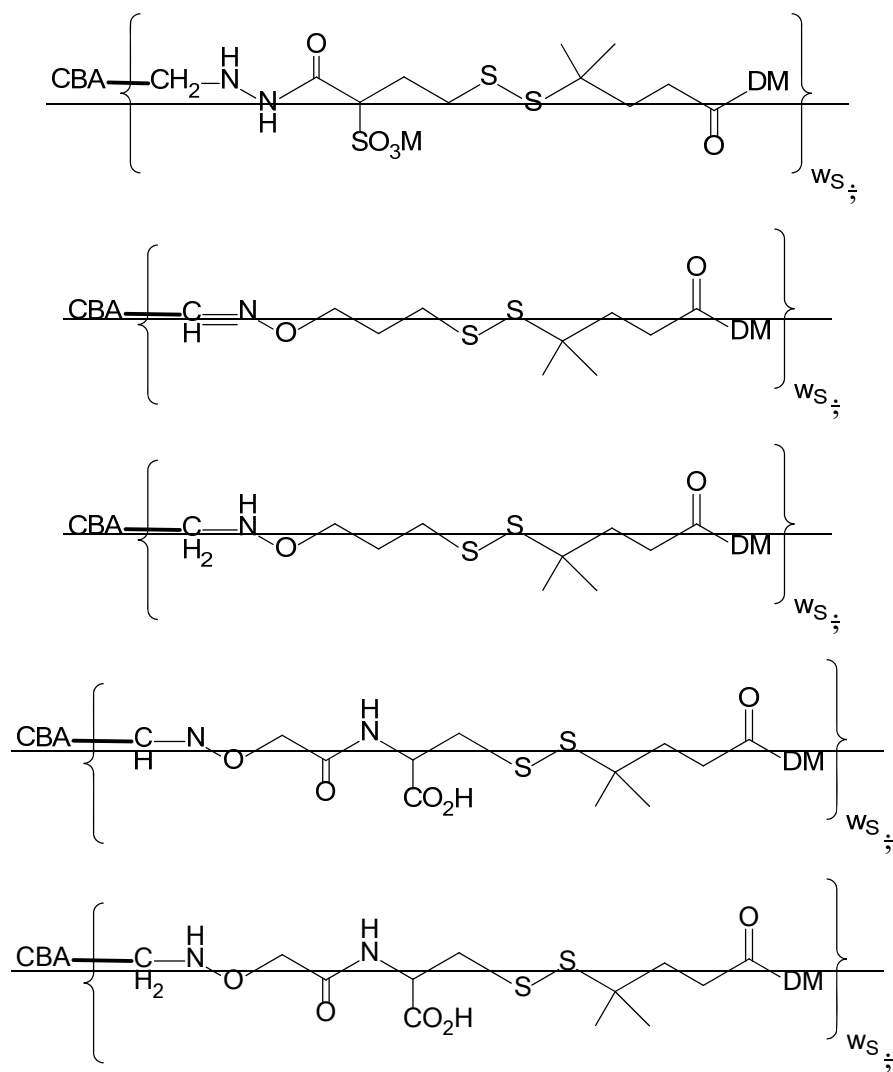
97. The immunoconjugate of any one of claims 94-96, wherein L<sub>1</sub> is represented by the following formula:



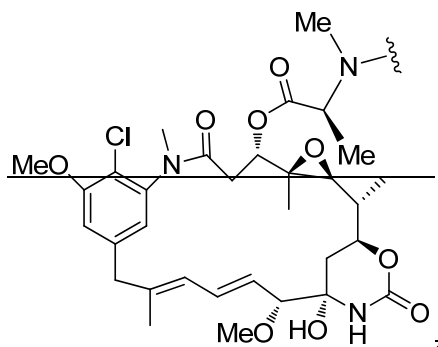
or a pharmaceutically acceptable salt thereof, wherein R is H or -SO<sub>3</sub>M and M is H<sup>+</sup> or a cation.

98. The immunoconjugate of claim 94, wherein the immunoconjugate is represented by the following formula:





or a pharmaceutically acceptable salt thereof; wherein DM is represented by the following formula:



99. An immunoconjugate having the following formula:



~~s3 is the site covalently linked to the group J<sub>CB</sub>' group;~~  
~~s4 is the site covalently linked to NMe group on Cy<sup>s4</sup>;~~  
~~Z<sub>b1</sub> and Z<sub>b2</sub> are both absent, or one of Z<sub>b1</sub> and Z<sub>b2</sub> is absent and the other is CH<sub>2</sub>-O or O-CH<sub>2</sub>;~~  
~~Z<sub>b1</sub>' and Z<sub>b2</sub>' are each independently absent, CH<sub>2</sub>-O, O-CH<sub>2</sub>, NR<sub>9</sub>-C(=O)-CH<sub>2</sub>, or CH<sub>2</sub>-C(=O)-NR<sub>9</sub>;~~  
~~R<sub>9</sub> is H or (C<sub>1</sub>-C<sub>3</sub>)alkyl;~~  
~~n1 and m1 are each independently an integer from 1 to 6;~~  
~~one of E<sub>1</sub> and E<sub>2</sub> is C(=O), and the other is NR<sub>9</sub>; or one of E<sub>1</sub> and E<sub>2</sub> is C(=O) or NR<sub>9</sub>, and the other is absent;~~  
~~P is an amino acid residue or a peptide containing between 2 to 20 amino acid residues; and~~  
~~R<sub>b1</sub>, R<sub>b2</sub>, R<sub>b3</sub>, R<sub>b4</sub>, R<sub>b5</sub> and R<sub>b6</sub>, for each occurrence, are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl.~~

100. ~~The immunoconjugate of claim 99, wherein R<sub>b1</sub>, R<sub>b2</sub>, R<sub>b3</sub>, R<sub>b4</sub>, R<sub>b5</sub>, and R<sub>b6</sub> are all H.~~

101. ~~The immunoconjugate of claim 100, wherein R<sub>9</sub> is H.~~

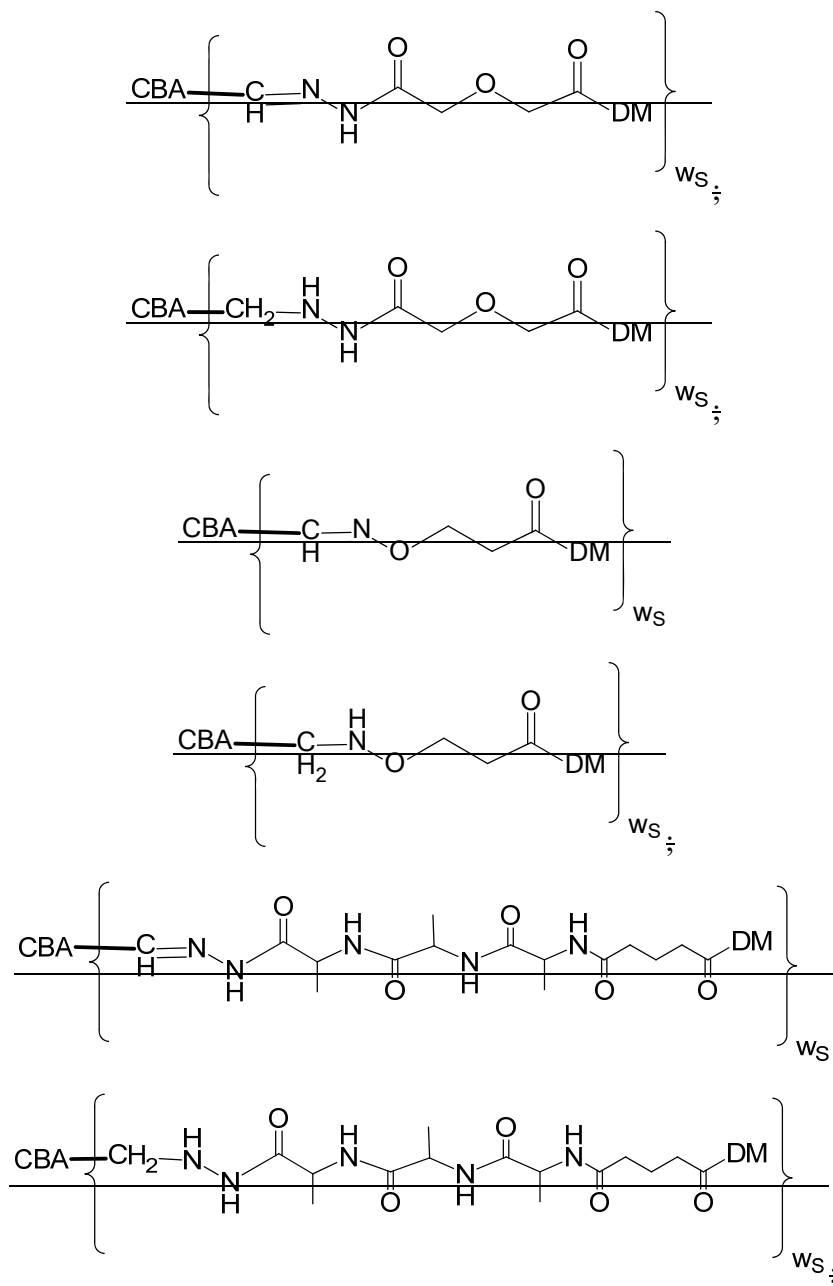
102. ~~The immunoconjugate of any one of claims 99-101, wherein Z<sub>b1</sub>' and Z<sub>b2</sub>' are both absent; or Z<sub>b1</sub>' is CH<sub>2</sub>-O; and Z<sub>b2</sub>' is absent; or Z<sub>b1</sub>' is CH<sub>2</sub>-C(=O)-NR<sub>9</sub>; and Z<sub>b2</sub>' is O-CH<sub>2</sub> or absent.~~

103. ~~The immunoconjugate of any one of claims 99-102, wherein P is a peptide containing 2 to 5 amino acid residues.~~

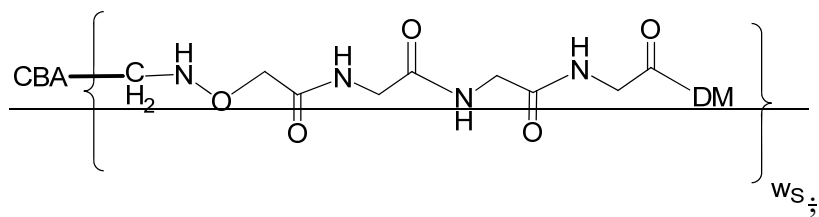
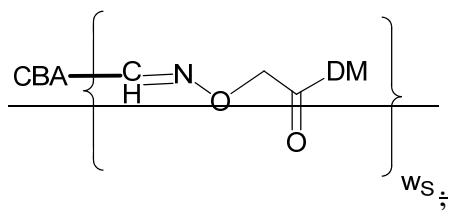
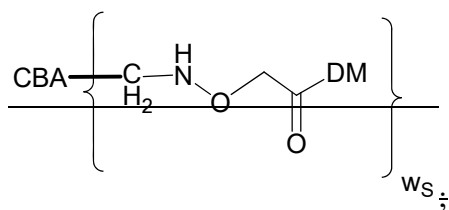
104. ~~The immunoconjugate of any one of claims 99-103, wherein P is selected from Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Ile-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl-Arg, Phe-N<sup>9</sup>-nitro-Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55), β-Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.~~

105. ~~The immunoconjugate of claim 104, wherein P is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, and D-Ala-D-Ala.~~

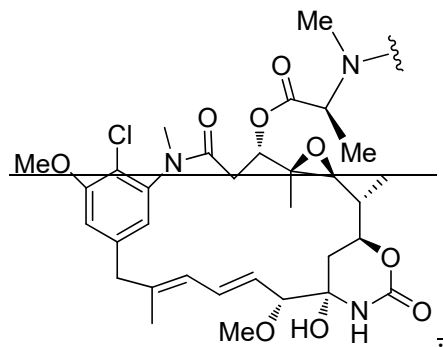
106. ~~The immunoconjugate of claim 99, wherein immunoconjugate is represented by the following formula:~~



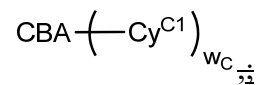




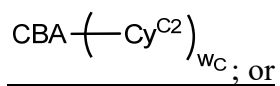
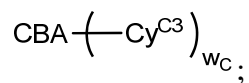
or a pharmaceutically acceptable salt thereof, wherein DM is represented by the following structural formula:



18 107. An immunoconjugate represented by the following formula:



;



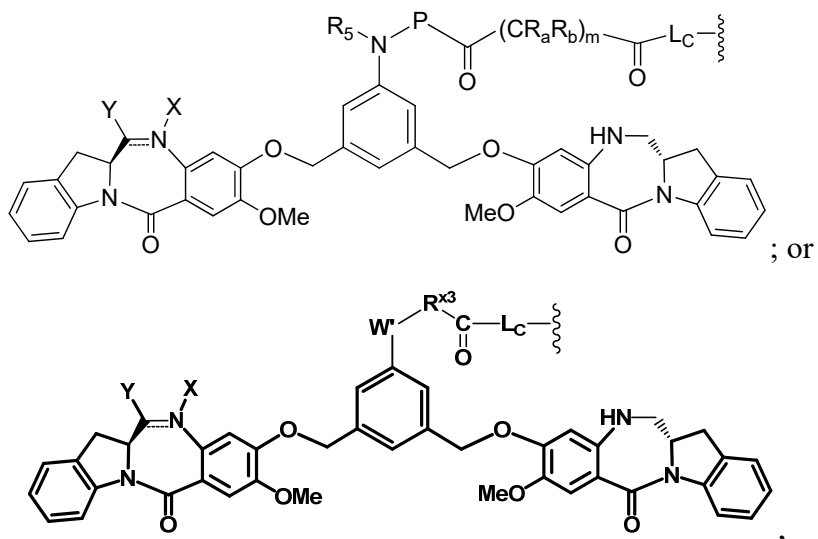
wherein:

CBA is an antibody or antigen-binding fragment thereof of claim 8 ~~any one of~~

~~claims 37, 29, 30, 38, 33, and 34, or the polypeptide thereof of claim 13 55 or 56,~~  
covalently linked to Cy<sup>C1</sup>, Cy<sup>C2</sup> or Cy<sup>C3</sup> through a cysteine residue;

W<sub>C</sub> is 1 or 2;

Cy<sup>C1</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line == between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is -H or an amine protecting moiety, Y is -OH or -SO<sub>3</sub>M, and M is H<sup>+</sup> or a cation;

R<sub>5</sub> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

P is an amino acid residue or a peptide containing 2 to 20 amino acid residues;

R<sub>a</sub> and R<sub>b</sub>, for each occurrence, are independently -H, (C<sub>1</sub>-C<sub>3</sub>)alkyl, or a charged substituent or an ionizable group Q;

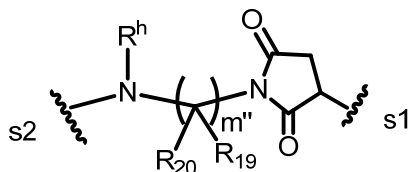
W' is -NR<sup>e'</sup>,

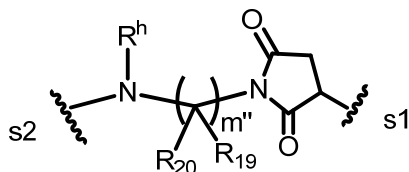
R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is -H or -Me;

R<sup>x3</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl; and,



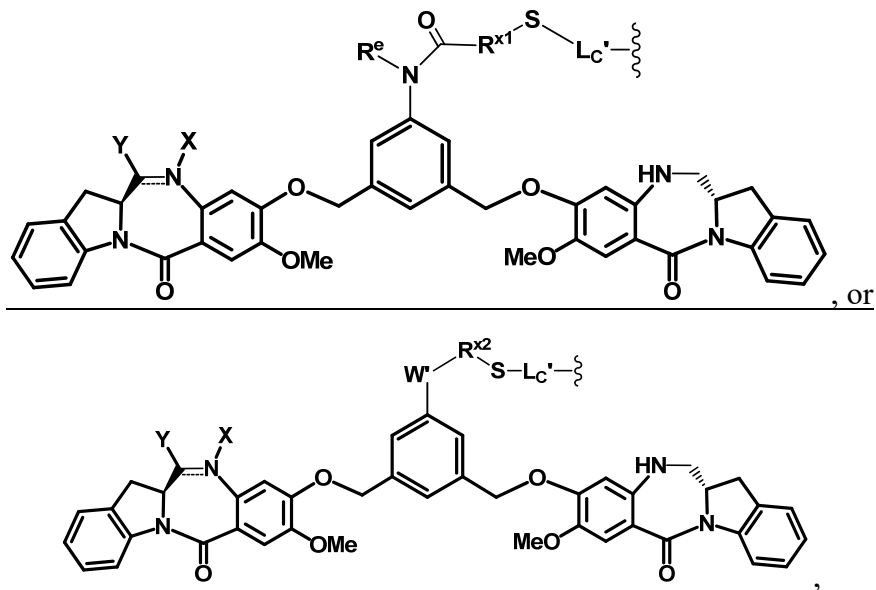
Lc is represented by , s1 is the site covalently linked to CBA, and s2 is the site covalently linked to the -C(=O)- group on Cy<sup>C1</sup>; wherein:

R<sub>19</sub> and R<sub>20</sub>, for each occurrence, are independently -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

m'' is an integer between 1 and 10; and

R<sup>h</sup> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Cy<sup>C2</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line  $\equiv$  between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is -H or an amine protecting moiety, Y is -OH or -SO<sub>3</sub>M, and M is H<sup>+</sup> or a cation;

R<sup>x1</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

R<sup>e</sup> is -H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

W' is -NR<sup>e'</sup>;

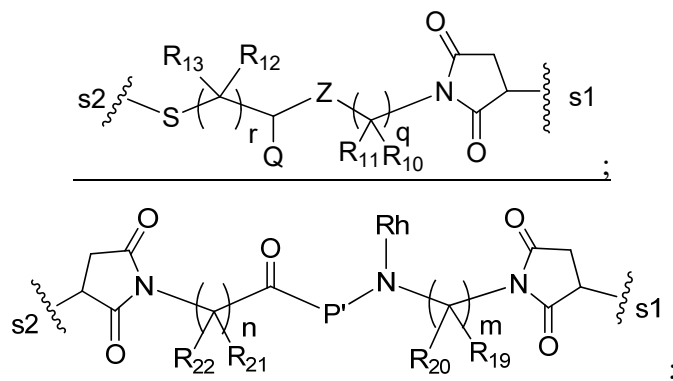
R<sup>e'</sup> is -(CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

$R^k$  is -H or -Me;

$R^{x2}$  is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

$L_C'$  is represented by the following formula:



wherein:

$s1$  is the site covalently linked to the CBA and  $s2$  is the site covalently linked to -S- group on Cy<sup>C2</sup>;

$Z$  is -C(=O)-NR<sub>9</sub>-, or -NR<sub>9</sub>-C(=O)-;

$Q$  is -H, a charged substituent, or an ionizable group;

$R_9$ ,  $R_{10}$ ,  $R_{11}$ ,  $R_{12}$ ,  $R_{13}$ ,  $R_{19}$ ,  $R_{20}$ ,  $R_{21}$  and  $R_{22}$ , for each occurrence, are independently -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

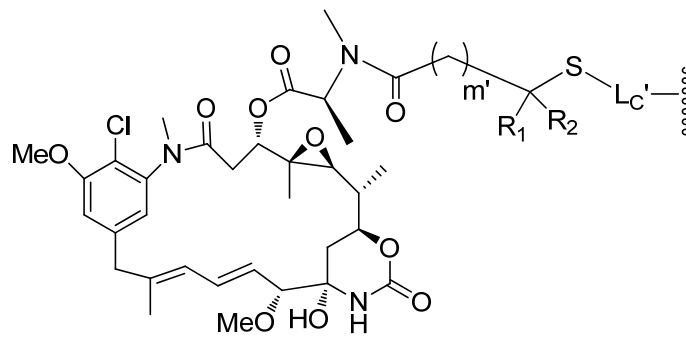
$q$  and  $r$ , for each occurrence, are independently an integer between 0 and 10;

$m$  and  $n$  are each independently an integer between 0 and 10;

$R^h$  is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

$P'$  is an amino acid residue or a peptide containing 2 to 20 amino acid residues;

Cy<sup>C3</sup> is represented by the following formula:

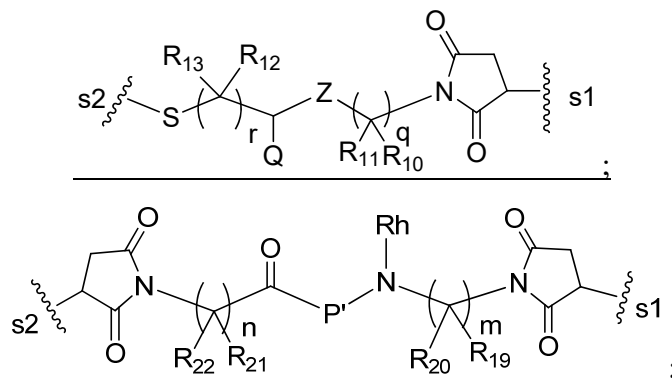


wherein:

$m'$  is 1 or 2;

R<sub>1</sub> and R<sub>2</sub>, are each independently -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

Lc' is represented by the following formula:



wherein:

s1 is the site covalently linked to the CBA and s2 is the site covalently linked to -S- group on Cy<sup>C3</sup>;

Z is -C(=O)-NR<sub>9</sub>-, or -NR<sub>9</sub>-C(=O)-;

Q is H, a charged substituent, or an ionizable group;

R<sub>9</sub>, R<sub>10</sub>, R<sub>11</sub>, R<sub>12</sub>, R<sub>13</sub>, R<sub>19</sub>, R<sub>20</sub>, R<sub>21</sub> and R<sub>22</sub>, for each occurrence, are independently -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

q and r, for each occurrence, are independently an integer between 0 and 10;

m and n are each independently an integer between 0 and 10;

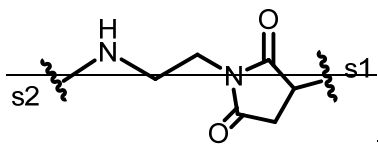
R<sup>h</sup> is -H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

P' is an amino acid residue or a peptide containing 2 to 20 amino acid residues.

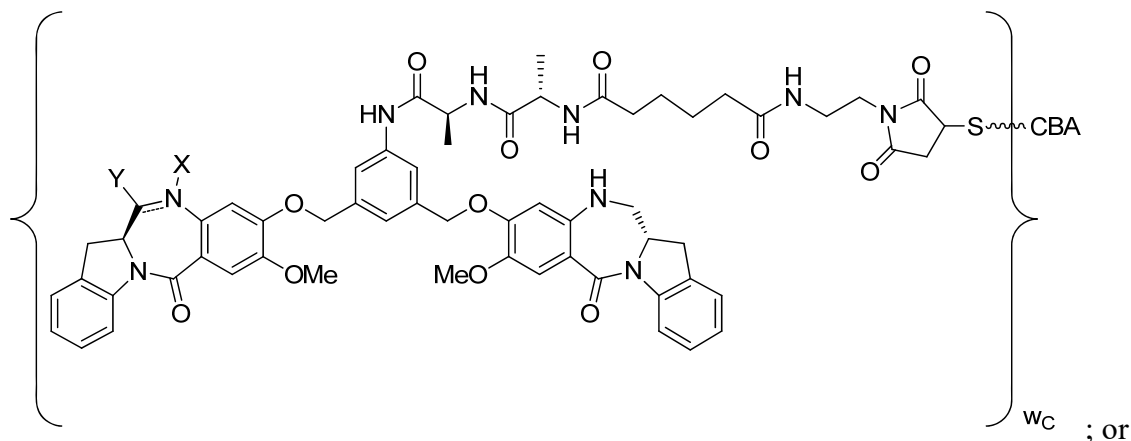
~~108. The immunoconjugate of claim 107, wherein R<sub>4</sub> and R<sub>5</sub> are both H; and R<sub>5</sub> is H or Me.~~

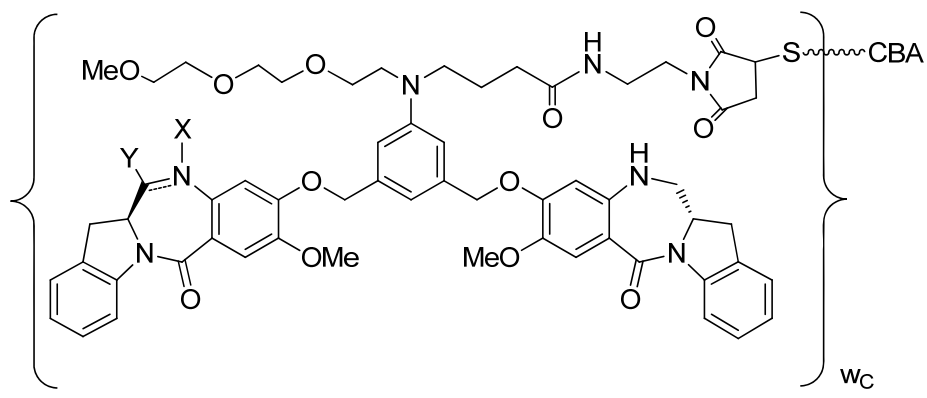
~~109. The immunoconjugate of claim 107 or 108, wherein P is a peptide containing 2 to 5 amino acid residues.~~

110. ~~The immunoconjugate of any one of claims 107-109, wherein P is selected from Gly-Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Ile-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl Arg, Phe-N<sup>9</sup>-nitro Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55),  $\beta$ -Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.~~
111. ~~The immunoconjugate of claim 110, wherein P is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, or D-Ala-D-Ala.~~
112. ~~The immunoconjugate of any one of claims 107-111, wherein Q is -SO<sub>3</sub>M.~~
113. ~~The immunoconjugate of any one of claims 107-112, wherein R<sub>19</sub> and R<sub>20</sub> are both H; and m'' is an integer from 1 to 6.~~
114. ~~The immunoconjugate of claim 113, wherein L<sub>C</sub> is represented by the following formula:~~



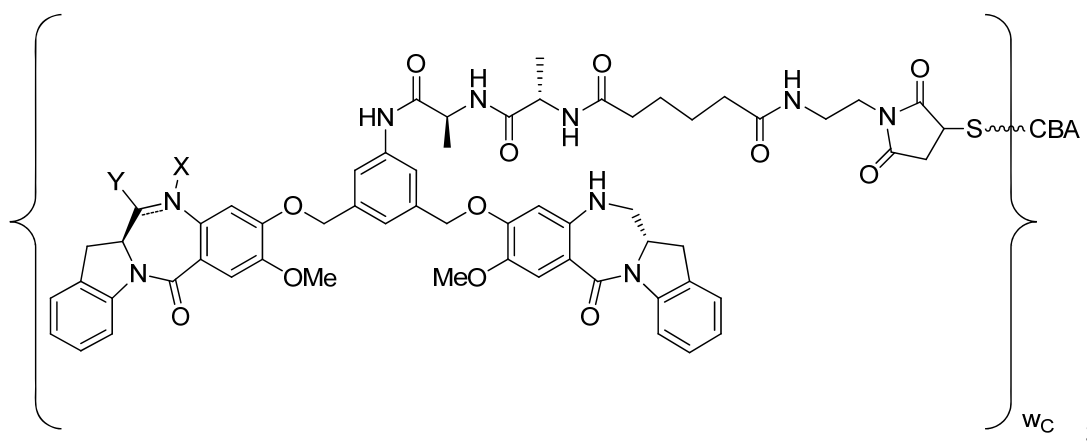
- ~~19~~ 115. The immunoconjugate of claim ~~18~~ 107, which is represented by the following formula:





or a pharmaceutically acceptable salt thereof, wherein the double line == between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H, and when it is a single bond, X is -H, and Y is -OH or -SO<sub>3</sub>M.

20. The immunoconjugate of claim 19, which is represented by the following formula:

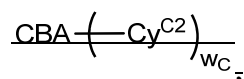


or a pharmaceutically acceptable salt thereof, wherein:

CBA is an antibody comprising an immunoglobulin heavy chain region having the amino acid sequence set forth in SEQ ID NO: 54; and an immunoglobulin light chain region having the amino acid sequence set forth in SEQ ID NO: 51; and

the double line == between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is -H, and when it is a single bond, X is -H, and Y is -OH or -SO<sub>3</sub>M.

116. An immunoconjugate represented by the following formula:

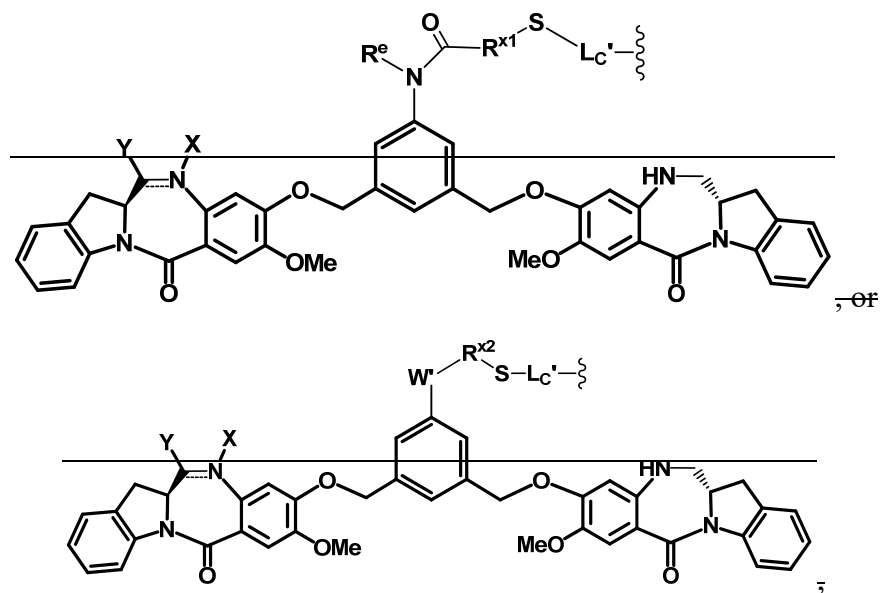


wherein:

CBA is an antibody or antigen-binding fragment thereof of any one of claims 37, 29, 30, 38, 33, and 34, or the polypeptide thereof of claim 55 or 56, covalently linked to Cy<sup>C2</sup> through a cysteine residue;

W<sub>C</sub> is 1 or 2;

Cy<sup>C2</sup> is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein:

the double line ~~==~~ between N and C represents a single bond or a double bond, provided that when it is a double bond, X is absent and Y is H or a (C<sub>1</sub>-C<sub>4</sub>)alkyl; and when it is a single bond, X is H or an amine protecting moiety, Y is OH or SO<sub>3</sub>M, and M is H<sup>+</sup> or a cation;

R<sup>x1</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

R<sup>e</sup> is H or a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

W' is NR<sup>e2</sup>;

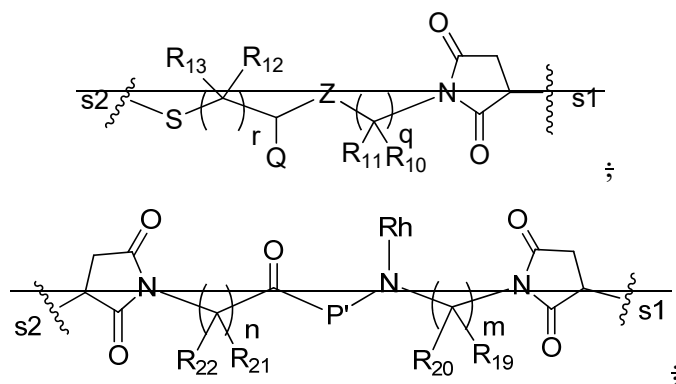
R<sup>e2</sup> is (CH<sub>2</sub>-CH<sub>2</sub>-O)<sub>n</sub>-R<sup>k</sup>;

n is an integer from 2 to 6;

R<sup>k</sup> is H or Me;

R<sup>x2</sup> is a (C<sub>1</sub>-C<sub>6</sub>)alkyl;

Lc' is represented by the following formula:



wherein:

s1 is the site covalently linked to the CBA and s2 is the site covalently linked to S-group on Cy<sup>62</sup>;

Z is  $\text{C}(=\text{O})\text{NR}_9$ , or  $\text{NR}_9\text{C}(=\text{O})$ ;

Q is H, a charged substituent, or an ionizable group;

$\text{R}_9$ ,  $\text{R}_{10}$ ,  $\text{R}_{11}$ ,  $\text{R}_{12}$ ,  $\text{R}_{13}$ ,  $\text{R}_{19}$ ,  $\text{R}_{20}$ ,  $\text{R}_{21}$  and  $\text{R}_{22}$ , for each occurrence, are independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;

q and r, for each occurrence, are independently an integer between 0 and 10;

m and n are each independently an integer between 0 and 10;

$\text{R}^h$  is H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and

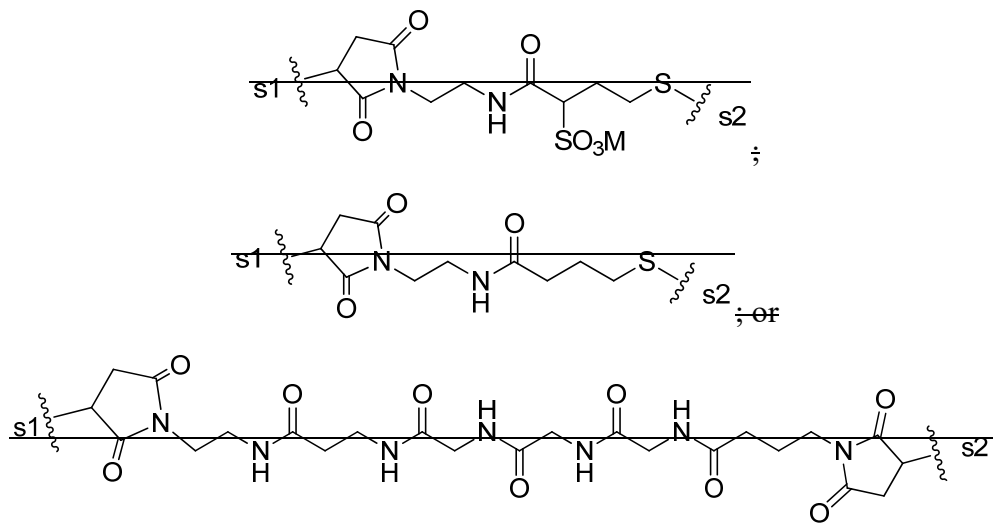
P' is an amino acid residue or a peptide containing 2 to 20 amino acid residues.

117. The immunoconjugate of claim 116, wherein P' is a peptide containing 2 to 5 amino acid residues.

118. The immunoconjugate of claim 116 or 117, wherein P' is selected from Gly-Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Ile-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl Arg, Phe-N<sup>9</sup>-nitro Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55),  $\beta$ -Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.

119. The immunoconjugate of claim 118, wherein P' is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, or D-Ala-D-Ala.

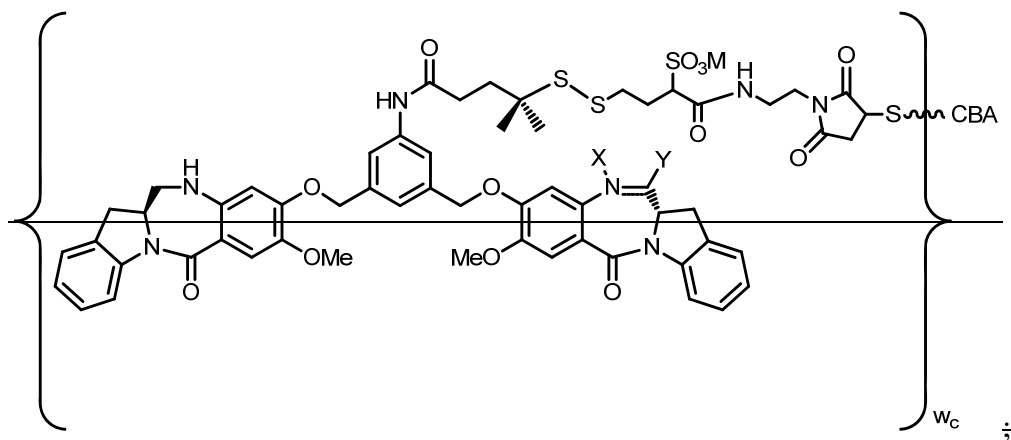
120. The immunoconjugate of claim 116, wherein  $L_c'$  is represented by the following formula:



121. The immunoconjugate of any one of claims 116-120,  $R^*$  is H or Me;  $R^{*1}$  is  $(CH_2)_p$ - $(CR^fR^*)$ , and  $R^{*2}$  is  $(CH_2)_p$ - $(CR^fR^*)$ , wherein  $R^f$  and  $R^*$  are each independently H or a  $(C_1-C_4)$ alkyl; and  $p$  is 0, 1, 2 or 3.

122. The immunoconjugate of claim 121, wherein  $R^f$  and  $R^*$  are the same or different, and are selected from H and Me.

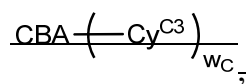
123. The immunoconjugate of claim 116, which is represented by the following formula:





126. ~~The immunoconjugate of claim 125, wherein M is H<sup>+</sup>, Na<sup>+</sup> or K<sup>+</sup>.~~

127. ~~An immunoconjugate having the following formula:~~

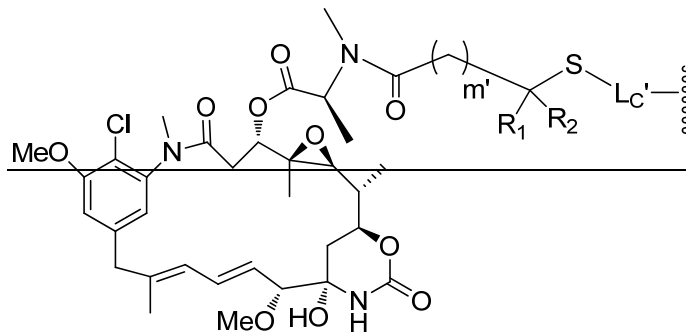


~~wherein:~~

~~CBA is an antibody or antigen binding fragment thereof of any one of claims 37, 29, 30, 38, 33, and 34, or the polypeptide thereof of claim 55 or 56, covalently linked to Cy<sup>C3</sup> through a cysteine residue;~~

~~w<sub>C</sub> is 1 or 2;~~

~~Cy<sup>C3</sup> is represented by the following formula:~~

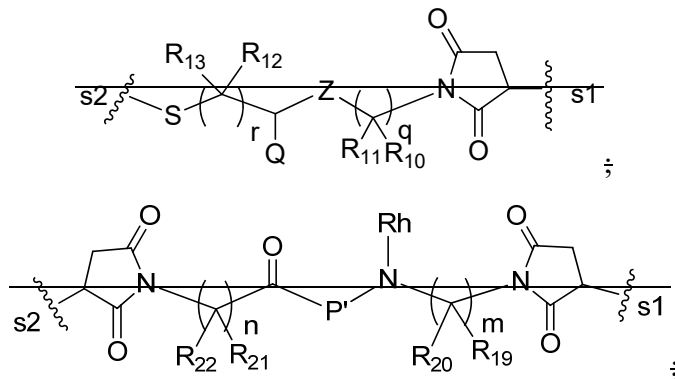


~~wherein:~~

~~m' is 1 or 2;~~

~~R<sub>1</sub> and R<sub>2</sub> are each independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;~~

~~Lc' is represented by the following formula:~~



~~wherein:~~

~~s1 is the site covalently linked to the CBA and s2 is the site covalently linked to S-group on Cy<sup>C3</sup>;~~

~~Z is C(=O)-NR<sub>9</sub>, or NR<sub>9</sub>-C(=O);~~

~~Q is H, a charged substituent, or an ionizable group;~~

~~R<sub>9</sub>, R<sub>10</sub>, R<sub>11</sub>, R<sub>12</sub>, R<sub>13</sub>, R<sub>19</sub>, R<sub>20</sub>, R<sub>21</sub> and R<sub>22</sub>, for each occurrence, are independently H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl;~~

~~q and r, for each occurrence, are independently an integer between 0 and 10;~~

~~m and n are each independently an integer between 0 and 10;~~

~~R<sup>h</sup> is H or a (C<sub>1</sub>-C<sub>3</sub>)alkyl; and~~

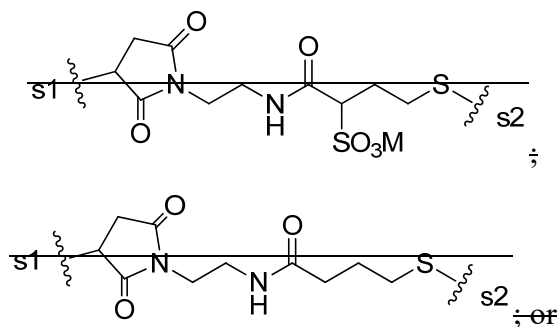
~~P' is an amino acid residue or a peptide containing 2 to 20 amino acid residues.~~

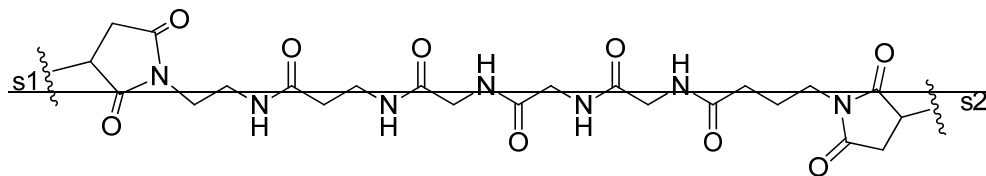
~~128. The immunoconjugate of claim 127, wherein P' is a peptide containing 2 to 5 amino acid residues.~~

~~129. The immunoconjugate of claim 127 or 128, wherein P' is selected from Gly-Gly-Gly, Ala-Val, Val-Ala, Val-Cit, Val-Lys, Phe-Lys, Lys-Lys, Ala-Lys, Phe-Cit, Leu-Cit, Lle-Cit, Trp, Cit, Phe-Ala, Phe-N<sup>9</sup>-tosyl Arg, Phe-N<sup>9</sup>-nitro Arg, Phe-Phe-Lys, D-Phe-Phe-Lys, Gly-Phe-Lys, Leu-Ala-Leu, Ile-Ala-Leu, Val-Ala-Val, Ala-Leu-Ala-Leu (SEQ ID NO: 55), β-Ala-Leu-Ala-Leu (SEQ ID NO: 57), Gly-Phe-Leu-Gly (SEQ ID NO: 73), Val-Arg, Arg-Val, Arg-Arg, Val-D-Cit, Val-D-Lys, Val-D-Arg, D-Val-Cit, D-Val-Lys, D-Val-Arg, D-Val-D-Cit, D-Val-D-Lys, D-Val-D-Arg, D-Arg-D-Arg, Ala-Ala, Ala-D-Ala, D-Ala-Ala, D-Ala-D-Ala, Ala-Met, and Met-Ala.~~

~~130. The immunoconjugate of claim 129, wherein P' is Gly-Gly-Gly, Ala-Val, Ala-Ala, Ala-D-Ala, D-Ala-Ala, or D-Ala-D-Ala.~~

~~131. The immunoconjugate of claim 127, wherein Lc' is represented by the following formula:~~



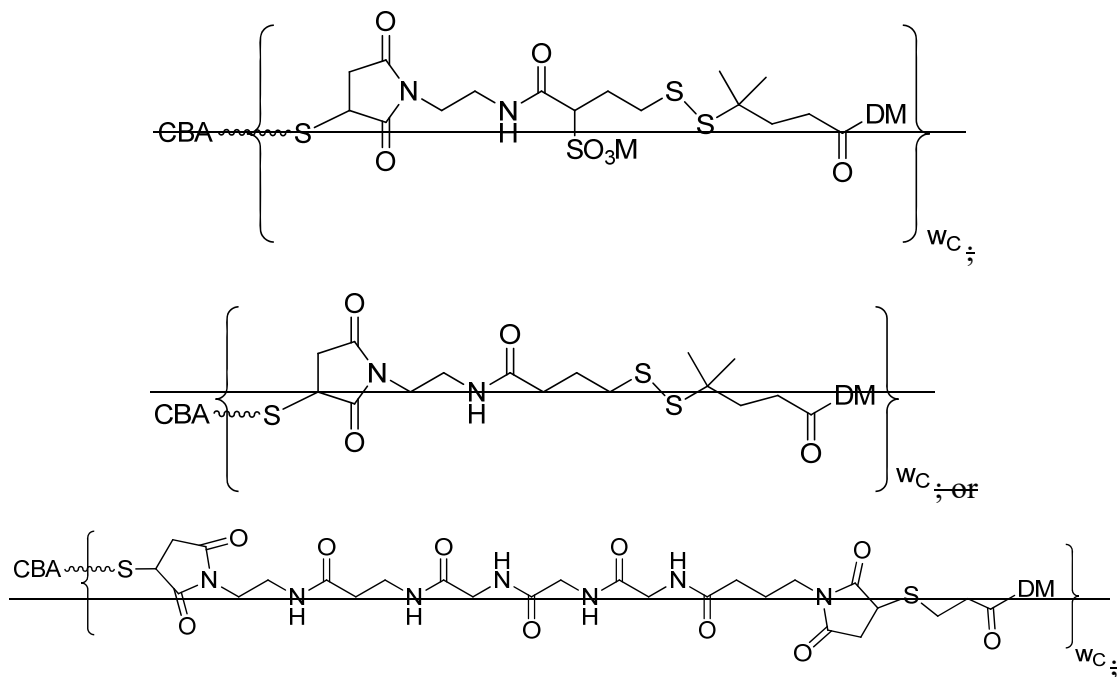


wherein M is H<sup>+</sup> or a cation.

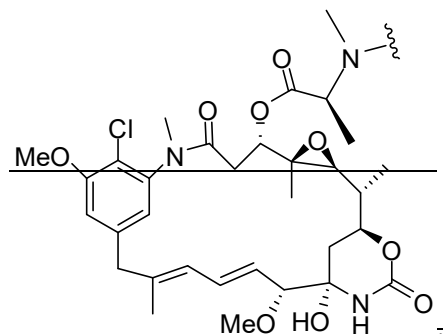
132. The immunoconjugate of any one of claims 127-131, wherein m' is 1 and R<sub>1</sub> and R<sub>2</sub> are both H.

133. The immunoconjugate of any one of claims 127-131, wherein m' is 2 and R<sub>1</sub> and R<sub>2</sub> are both Me.

134. The immunoconjugate of claim 127, wherein the immunoconjugate is represented by the following formula:



or a pharmaceutically acceptable salt thereof, wherein DM is a drug moiety represented by the following formula:



21 ~~135~~. A pharmaceutical composition comprising the antibody or antigen-binding fragment thereof of any one of claims 1-12 ~~1-54~~, or the polypeptide of claim 13 ~~55 or 56~~, or the immunoconjugate of any one of claims 14-20 ~~60-134~~, and a pharmaceutically acceptable carrier.

22 ~~136~~. A method for inhibiting the growth of a cell expressing CD123, comprising contacting the cell with the antibody or antigen-binding fragment thereof of any one of claims 1-12 ~~1-54~~, or the polypeptide of claim 13 ~~55 or 56~~, or the immunoconjugate of any one of claims 14-20 ~~60-134~~, or the pharmaceutical composition of claim 21 ~~135~~.

~~137. The method of claim 136, wherein said cell is a tumor cell.~~

~~138. The method of claim 137, wherein said cell is a leukemia cell or a lymphoma cell.~~

23~~139~~. Use of the antibody or antigen-binding fragment thereof any one of claims 1-12, or the polypeptide of claim 13, or the immunoconjugate of any one of claims 14-20 in the manufacture of a medicament~~A method for treating a subject having cancer, wherein cells of the cancer expresses CD123, the method comprising administering to said subject a therapeutically effective amount of the antibody or antigen-binding fragment thereof of any one of claims 1-54, or the polypeptide of claim 55 or 56, or the immunoconjugate of any one of claims 60-134, or the pharmaceutical composition of claim 135.~~

24~~140~~. The ~~method~~ use of claim 23~~139~~, wherein said cancer is leukemia or lymphoma.

25~~141~~. The ~~method~~ use of claim 23~~139~~, wherein said cancer is selected from the group consisting of: acute myeloid leukemia (AML), acute lymphoblastic leukemia (ALL), and Chronic lymphocytic leukemia (CLL).

26~~142~~. The ~~method~~ use of claim 25~~141~~, wherein said cancer is acute myeloid leukemia (AML).

27143. The ~~method~~ use of claim 25141, wherein said cancer is B-cell acute lymphoblastic leukemia (B-ALL).

28144. Use of the antibody or antigen-binding fragment thereof any one of claims 1-12, or the polypeptide of claim 13, or the immunoconjugate of any one of claims 14-20 in the manufacture of a medicament~~A method~~ for treating a cell-proliferative disorder in a subject, wherein cells of the cell-proliferative disorder expresses CD123,~~the method comprising administering to said subject a therapeutically effective amount of the antibody or antigen-binding fragment thereof of any one of claims 1-54, or the polypeptide of claim 55 or 56, or the immunoconjugate of any one of claims 60-134, or the pharmaceutical composition of claim 135, in an amount sufficient to treat said cell-proliferative disorder.~~

29145. The ~~use~~method of claim 28144, wherein said cell-proliferative disorder is selected from the group consisting of: acute myeloid leukemia (AML), chronic myeloid leukemia (CML), acute lymphoblastic leukemia (ALL), B-cell lineage acute lymphoblastic leukemia (B-ALL), chronic lymphocytic leukemia (CLL), hairy cell leukemia (HCL), myelodysplastic syndrome, basic plasmacytoid DC neoplasm (BPDCN) leukemia, non-Hodgkin lymphomas (NHL), mantle cell lymphoma, and Hodgkin's leukemia (HL).