

CLAIMS

1. An antibody or antigen binding fragment thereof that binds to human CCR7 protein, comprising:
 - a. a heavy chain variable region that comprises an HCDR1 (Heavy Chain Complementarity Determining Region 1) of SEQ ID NO:1, an HCDR2 (Heavy Chain Complementarity Determining Region 2) of SEQ ID NO:2, and an HCDR3 (Heavy Chain Complementarity Determining Region 3) of SEQ ID NO:3; and a light chain variable region that comprises an LCDR1 (Light Chain Complementarity Determining Region 1) of SEQ ID NO:17, an LCDR2 (Light Chain Complementarity Determining Region 2) of SEQ ID NO:18, and an LCDR3 (Light Chain Complementarity Determining Region 3) of SEQ ID NO:19;
 - b. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:4, an HCDR2 of SEQ ID NO:5, and an HCDR3 of SEQ ID NO:6; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:20, an LCDR2 of SEQ ID NO:21, and an LCDR3 of SEQ ID NO:22;
 - c. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:7, an HCDR2 of SEQ ID NO:8, and an HCDR3 of SEQ ID NO:9; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:23, an LCDR2 of SEQ ID NO:24, and an LCDR3 of SEQ ID NO:25;
 - d. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:10, an HCDR2 of SEQ ID NO:11, and an HCDR3 of SEQ ID NO:12; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:26, an LCDR2 of SEQ ID NO:27, and an LCDR3 of SEQ ID NO:28;
 - e. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:33, an HCDR2 of SEQ ID NO:34, and an HCDR3 of SEQ ID NO:35; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:49, an LCDR2 of SEQ ID NO:50, and an LCDR3 of SEQ ID NO:51;

- f. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:36, an HCDR2 of SEQ ID NO:37, and an HCDR3 of SEQ ID NO:38; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:52, an LCDR2 of SEQ ID NO:53, and an LCDR3 of SEQ ID NO:54;
- g. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:39, an HCDR2 of SEQ ID NO:40, and an HCDR3 of SEQ ID NO:41; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:55, an LCDR2 of SEQ ID NO:56, and an LCDR3 of SEQ ID NO:57;
- h. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:42, an HCDR2 of SEQ ID NO:43, and an HCDR3 of SEQ ID NO:44; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:58, an LCDR2 of SEQ ID NO:59, and an LCDR3 of SEQ ID NO:60;
- i. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:65, an HCDR2 of SEQ ID NO:66, and an HCDR3 of SEQ ID NO:67; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:81, an LCDR2 of SEQ ID NO:82, and an LCDR3 of SEQ ID NO:83;
- j. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:68, an HCDR2 of SEQ ID NO:69, and an HCDR3 of SEQ ID NO:70; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:84, an LCDR2 of SEQ ID NO:85, and an LCDR3 of SEQ ID NO:86;
- k. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:71, an HCDR2 of SEQ ID NO:72, and an HCDR3 of SEQ ID NO:73; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:87, an LCDR2 of SEQ ID NO:88, and an LCDR3 of SEQ ID NO:89;
- l. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:74, an HCDR2 of SEQ ID NO:75, and an HCDR3 of SEQ ID NO:76; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:90, an LCDR2 of SEQ ID NO:91, and an LCDR3 of SEQ ID NO:92;

- m. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:596, an HCDR2 of SEQ ID NO:597, and an HCDR3 of SEQ ID NO:598; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:612, an LCDR2 of SEQ ID NO:613, and an LCDR3 of SEQ ID NO:614;
 - n. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:599, an HCDR2 of SEQ ID NO:600, and an HCDR3 of SEQ ID NO:601; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:615, an LCDR2 of SEQ ID NO:616, and an LCDR3 of SEQ ID NO:617;
 - o. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:602, an HCDR2 of SEQ ID NO:603, and an HCDR3 of SEQ ID NO:604; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:618, an LCDR2 of SEQ ID NO:619, and an LCDR3 of SEQ ID NO:620; or
 - p. a heavy chain variable region that comprises an HCDR1 of SEQ ID NO:605, an HCDR2 of SEQ ID NO:606, and an HCDR3 of SEQ ID NO:607; and a light chain variable region that comprises an LCDR1 of SEQ ID NO:621, an LCDR2 of SEQ ID NO:622, and an LCDR3 of SEQ ID NO:623.
2. The antibody of claim 1, wherein the antibody or antigen binding fragment thereof has reduced or no significant effector function as compared to a wild-type antibody of the same isotype.
 3. The antibody of claim 2, wherein the antibody or antigen binding fragment thereof has a reduced level of, or no significant level of antibody-dependent cell-mediated cytotoxicity (ADCC) activity.
 4. The antibody of any one of claims 1-3, wherein the antibody or antigen binding fragment thereof comprises a silenced Fc region.
 5. The antibody of claim 4, wherein the antibody or antigen binding fragment thereof

comprises a mutation in the Fc region selected from: D265A; P329A; P329G; N297A; D265A and P329A; D265A and N297A; L234 and L235A; P329A, L234A and L235A; and P329G, L234A and L235A.

6. The antibody of any one of claims 1-5, wherein the antibody or antigen binding fragment thereof has no significant cell killing activity.

7. The antibody of any one of claims 1-6, wherein the antibody or antigen binding fragment thereof binds with greater affinity to cells expressing higher levels of CCR7 than cells expressing lower levels of CCR7.

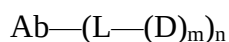
8. The antibody of claim 7, wherein the antibody or antigen binding fragment thereof binds with greater affinity to cancer cells that express higher levels of CCR7 than normal cells that express lower level of CCR7.

9. The antibody of any one of claims 1-8, wherein the antibody or antigen binding fragment thereof does not significantly deplete normal hematopoietic cells that express CCR7.

10. An antibody or antigen binding fragment thereof that binds CCR7 comprising:

- a. A heavy chain variable region (VH) comprising the amino acid sequence of SEQ ID NO:13, and a light chain variable region (VL) comprising the amino acid sequence of SEQ ID NO:29;
- b. A heavy chain variable region (VH) comprising the amino acid sequence of SEQ ID NO:45, and a light chain variable region (VL) comprising the amino acid sequence of SEQ ID NO:61;
- c. A heavy chain variable region (VH) comprising the amino acid sequence of SEQ ID NO:77, and a light chain variable region (VL) comprising the amino acid sequence of SEQ ID NO:93; or

- d. A heavy chain variable region (VH) comprising the amino acid sequence of SEQ ID NO:608, and a light chain variable region (VL) comprising the amino acid sequence of SEQ ID NO:624.
11. An antibody or antigen binding fragment thereof that binds CCR7 comprising:
- a. A heavy chain comprising the amino acid sequence of SEQ ID NO:15, and a light chain comprising the amino acid sequence of SEQ ID NO:31;
 - b. A heavy chain comprising the amino acid sequence of SEQ ID NO:47, and a light chain comprising the amino acid sequence of SEQ ID NO:63;
 - c. A heavy chain comprising the amino acid sequence of SEQ ID NO:79, and a light chain comprising the amino acid sequence of SEQ ID NO:95; or
 - d. A heavy chain comprising the amino acid sequence of SEQ ID NO:610, and a light chain comprising the amino acid sequence of SEQ ID NO:626.
12. The antibody of any one of claims 1-11, wherein the antibody or antigen binding fragment thereof comprises one or more cysteine substitutions.
13. The antibody of claim 12, wherein the antibody or antigen binding fragment thereof comprises one or more cysteine substitutions selected from S152C, S375C, or both S152C and S375C of the heavy chain of the antibody or antigen binding fragment thereof, wherein the position is numbered according to the EU system.
14. The antibody of any one of claims 1-13, wherein the antibody is a monoclonal antibody.
15. An antibody drug conjugate comprising the formula



or a pharmaceutically acceptable salt thereof; wherein

Ab is an antibody or antigen binding antigen binding fragment thereof according to any one of claims 1 to 14;

L is a linker;

D is a drug moiety;

m is an integer from 1 to 8; and

n is an integer from 1 to 12.

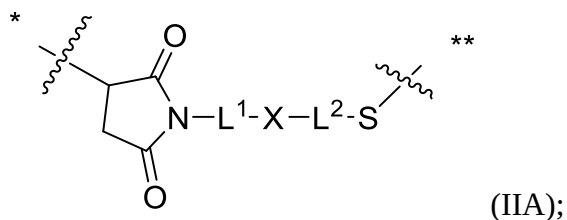
16. The antibody drug conjugate of claim 15, wherein m is 1.

17. The antibody drug conjugate of any one of claims 15 or 16, wherein n is about 3 to about 4.

18. The antibody drug conjugate of any one of claims 15-17, wherein the linker is selected from the group consisting of a cleavable linker, a non-cleavable linker, a hydrophilic linker, a procharged linker, and a dicarboxylic acid based linker.

19. The antibody drug conjugate of claim 18, wherein the linker is derived from a cross-linking reagent selected from the group consisting of N-succinimidyl-3-(2-pyridyldithio)propionate (SPDP), N-succinimidyl 4-(2-pyridyldithio)pentanoate (SPP), N-succinimidyl 4-(2-pyridyldithio)butanoate (SPDB), N-succinimidyl-4-(2-pyridyldithio)-2-sulfo-butanoate (sulfo-SPDB), N-succinimidyl iodoacetate (SIA), N-succinimidyl(4-iodoacetyl)aminobenzoate (SIAB), maleimide PEG NHS, N-succinimidyl 4-(maleimidomethyl) cyclohexanecarboxylate (SMCC), N- sulfosuccinimidyl 4-(maleimidomethyl) cyclohexanecarboxylate (sulfo-SMCC), and 2,5-dioxopyrrolidin-1-yl 17-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)-5,8,11,14-tetraoxo-4,7,10,13-tetraazaheptadecan-1-oate (CX1-1).

20. The antibody drug conjugate of claim 18, wherein the linker has the following Formula (IIA):



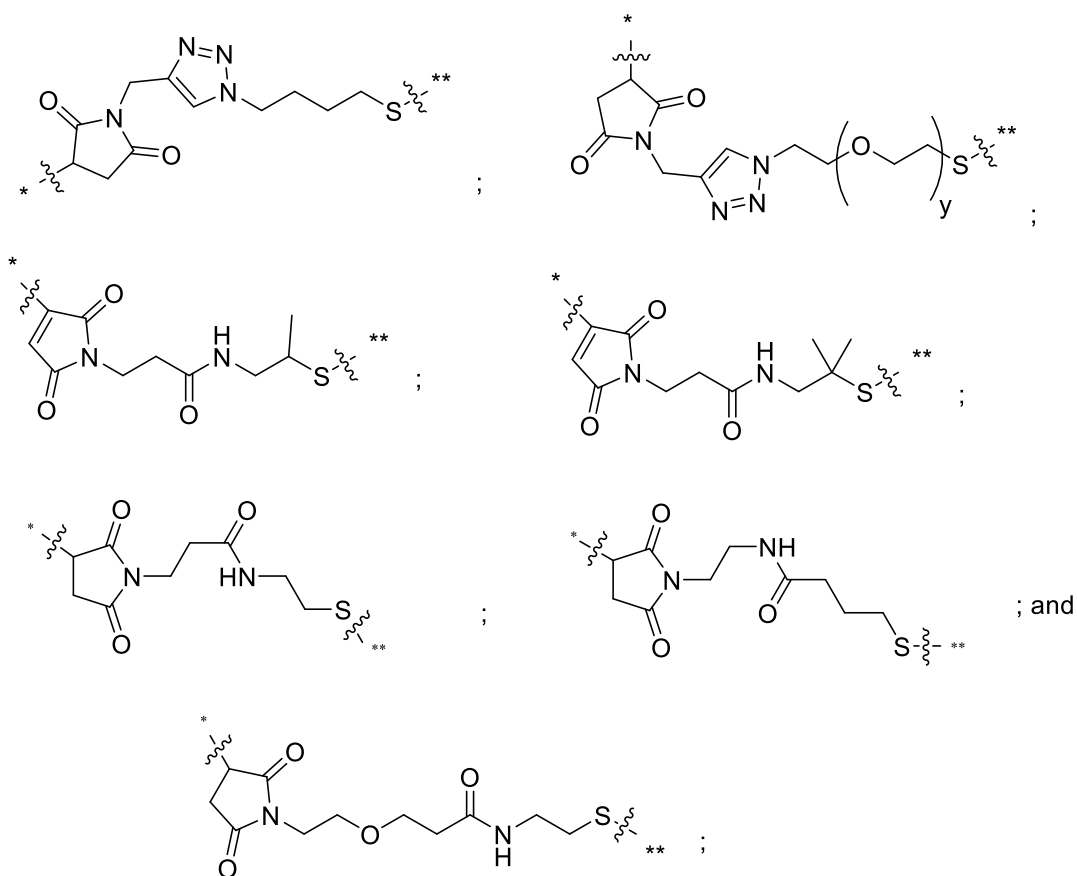
wherein * is linked to the thiol functionality on the antibody, and ** is linked to the thiol functionality of a drug moiety; and wherein:

L^1 is a C_{1-6} alkylene wherein one of the methylene groups may be replaced with oxygen;

L^2 is a C_{1-6} alkylene or is $-(CH_2CH_2O)_y-CH_2-CH_2-$ wherein y is 1 to 11;

X is $-C(O)-NH-$, $-NHC(O)-$ or a triazole; and alkylene is linear or branched.

21. The antibody drug conjugate of claim 20 wherein the linker has the following Formula:



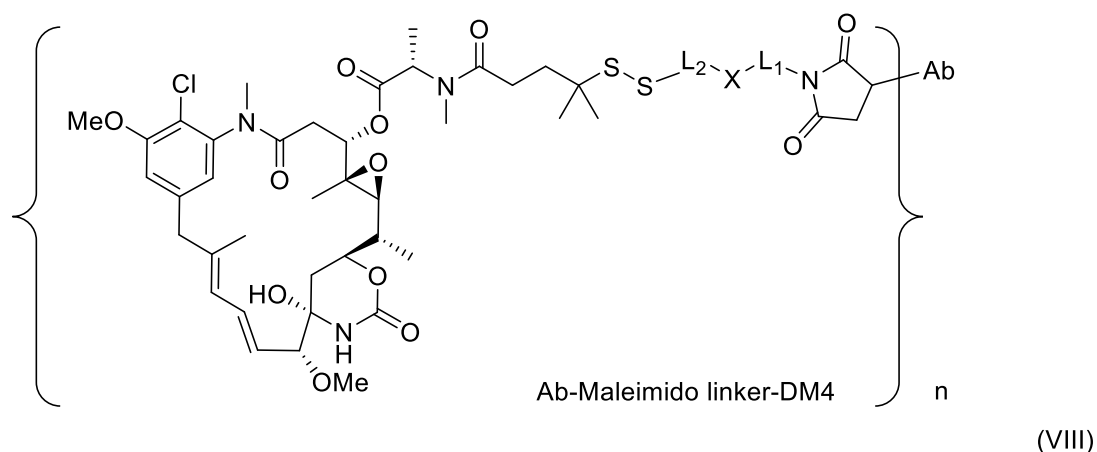
wherein y is 1 to 11; * is linked to the thiol functionality on the antibody, and ** is linked to the thiol functionality of the drug moiety.

22. The antibody drug conjugate of any one of claims 15-21, wherein the drug moiety is selected from a group consisting of a V-ATPase inhibitor, a pro-apoptotic agent, a Bcl2 inhibitor, an MCL1 inhibitor, a HSP90 inhibitor, an IAP inhibitor, an mTor inhibitor, a microtubule stabilizer, a microtubule destabilizer, an auristatin, a dolastatin, a maytansinoid, a MetAP (methionine aminopeptidase), an auristatin, an amanitin, a pyrrolobenzodiazepine, an RNA polymerase inhibitor, an inhibitor of nuclear export of proteins CRM1, a DPPIV inhibitor, proteasome inhibitors, inhibitors of phosphoryl transfer reactions in mitochondria, a protein synthesis inhibitor, a kinase inhibitor, a CDK2 inhibitor, a CDK9 inhibitor, a kinesin inhibitor, an HDAC inhibitor, a DNA damaging agent, a DNA alkylating agent, a DNA intercalator, a DNA minor groove binder and a DHFR inhibitor.

23. The antibody drug conjugate of claim 22, wherein the cytotoxic agent is a maytansinoid.

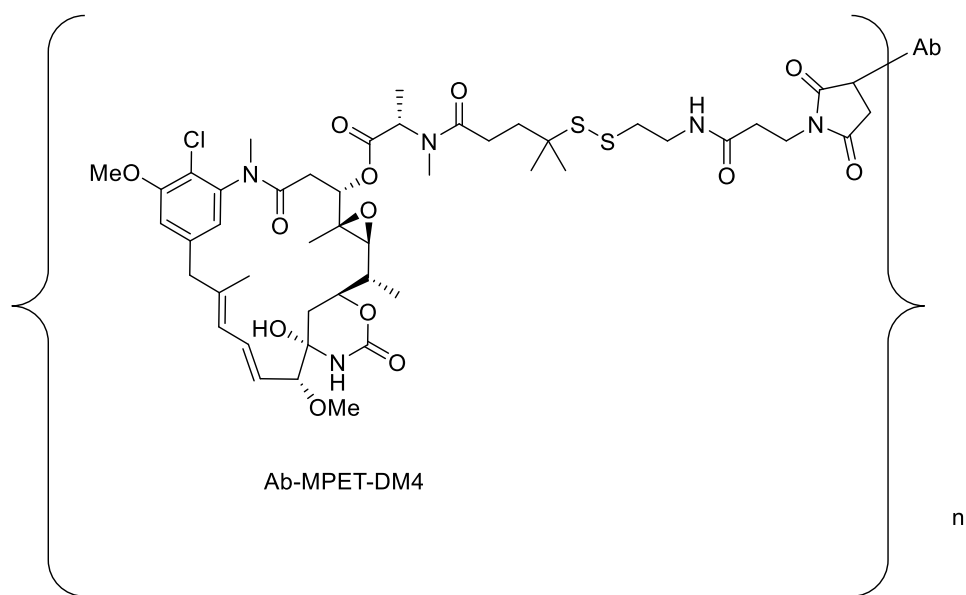
24. The antibody drug conjugate of claim 23, wherein the maytansinoid is N(2')- deacetyl-N(2')-(3-mercapto-1-oxopropyl)-maytansine (DM1), N(2')-deacetyl-N(2')-(4-mercapto-1-oxopentyl)-maytansine (DM3) or N(2')-deacetyl-N2-(4- mercapto-4-methyl- 1 -oxopentyl)-maytansine (DM4).

25. The antibody drug conjugate of any one of claims 15-24 having the following formula (VIII):



wherein L^1 is a C_{1-6} alkylene wherein one of the methylene groups may be replaced with oxygen;
 L^2 is a C_{1-6} alkylene or is $-(CH_2CH_2O)_y-CH_2-CH_2-$ wherein y is 1 to 11;
 X is $-C(O)-NH-$, $-NHC(O)-$ or a triazole; and alkylene is linear or branched; and wherein n is about 3 to about 4; or a pharmaceutically acceptable salt thereof

26. An antibody drug conjugate having the following formula:



wherein n is about 3 to about 4, and Ab is an antibody comprising a heavy chain comprising the amino acid sequence of SEQ ID NO:47, and a light chain comprising the amino acid sequence of SEQ ID NO:63; or a pharmaceutically acceptable salt thereof.

27. A pharmaceutical composition comprising the antibody, or antigen binding fragment thereof, of any one of claims 1-14 and a pharmaceutically acceptable carrier.

28. A pharmaceutical composition comprising the antibody drug conjugate of any one of claims 15-26 and a pharmaceutically acceptable carrier.

29. Use of the antibody drug conjugate of any one of claims 15-26, or the pharmaceutical composition of claims 27 or 28, in the manufacture of a medicament for the treatment or prevention of cancer in a patient in need thereof, wherein the cancer is a CCR7 expressing cancer.

30. The use of claim 29, wherein the medicament is to be administered to the patient in combination with one or more additional therapeutic compounds.

31. The use of claim 30, wherein the one or more additional therapeutic compounds is selected from a standard of care chemotherapeutic, a costimulatory molecule, or a checkpoint inhibitor.

32. The use of claim 31, wherein the costimulatory molecule is selected from an agonist of OX40, CD2, CD27, CD28, ICAM-1, LFA-1 (CD11a/CD18), ICOS (CD278), 4-1BB (CD137), GITR, CD30, CD40, BAFFR, HVEM, CD7, LIGHT, NKG2C, SLAMF7, NKp80, CD160, B7-H3, STING, or CD83 ligand.

33. The use of claim 31, wherein the checkpoint inhibitor is selected from an inhibitor of PD-1, PD-L1, PD-L2, CTLA4, TIM3, LAG3, VISTA, BTLA, TIGIT, LAIR1, CD160, 2B4 and/or TIGIT beta.

34. Use of the antibody or antigen binding fragment thereof of any one of claims 1-14, in the manufacture of a medicament to treat or prevent a CCR7 expressing cancer in a patient in need thereof.

35. Use of the pharmaceutical composition of any one of claims 27 or 28 in the manufacture of a medicament for the treatment or prevention of cancer in a patient in need thereof, a CCR7 expressing cancer.

36. The use of any one of claims 29 to 35, wherein the cancer expresses CCR7.

37. The use of claim 36, wherein the cancer is selected from the group consisting of chronic lymphocytic leukemia (CLL), peripheral T cell lymphomas (PTCL) such as adult T-cell leukemia/lymphoma (ATLL) and anaplastic large-cell lymphoma (ALCL), Non-Hodgkin's lymphoma (NHL) such as mantle cell lymphoma (MCL), Burkitt's lymphoma and diffuse large B-cell lymphoma (DLBCL), gastric carcinoma, non-small cell lung cancer, small cell lung cancer, head and neck cancer, nasopharyngeal carcinoma (NPC), esophageal cancer, colorectal carcinoma, pancreatic cancer, thyroid cancer, breast cancer, renal cell cancer, and cervical cancer.

38. The use of claim 37, wherein the cancer is selected from the group consisting of chronic lymphocytic leukemia (CLL), peripheral T cell lymphomas (PTCL) such as adult T-cell leukemia/lymphoma (ATLL) and anaplastic large-cell lymphoma (ALCL), Non-Hodgkin's lymphoma (NHL) such as mantle cell lymphoma (MCL), Burkitt's lymphoma and diffuse large B-cell lymphoma (DLBCL), and non-small cell lung cancer.

39. A nucleic acid that encodes the antibody or antigen binding fragment of any one of claims 1-14.

40. The nucleic acid of claim 39, wherein the nucleic acid comprises the nucleotide sequence of SEQ ID NOs: 14, 16, 30, 32, 46, 48, 62, 64, 78, 80, 94, 96, 481, 483, 497, or 499.

41. A vector comprising the nucleic acid of claim 39 or 40.

42. A host cell comprising the vector according to claim 41, or the nucleic acid according to claim 39 or 40.

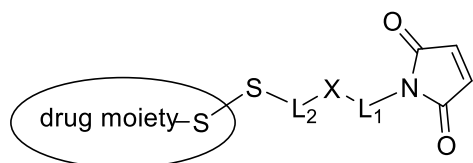
43. A process for producing an antibody or antigen binding fragment comprising cultivating the host cell of claim 42 and recovering the antibody from cell culture.

44. The process of claim 43 wherein recovering the antibody from cell culture comprises the steps of:

- a) removing cells and filtering the culture;
- b) purifying the culture by affinity chromatography ;
- c) inactivating any viruses in the culture by adjusting the pH to 3.4-3.6, then readjusting the pH to 5.8-6.2 and filtering the culture;
- d) purifying the culture by cation exchange chromatography and performing on-column reduction of the culture;
- e) performing anion exchange chromatography on the culture;
- f) removing viruses by nanofiltration;
- g) filtering the culture containing the antibody; and
- h) obtaining purified antibody.

45. A process for producing an anti-CCR7 antibody drug conjugate comprising:

- (a) pre-forming a linker-drug moiety of the following Formula:



wherein:

the drug moiety is DM1, DM3 or DM4 and the drug moiety is attached to the linker via its thiol functionality;

L¹ is a C₁₋₆alkylene wherein one of the methylene groups may be replaced with oxygen;

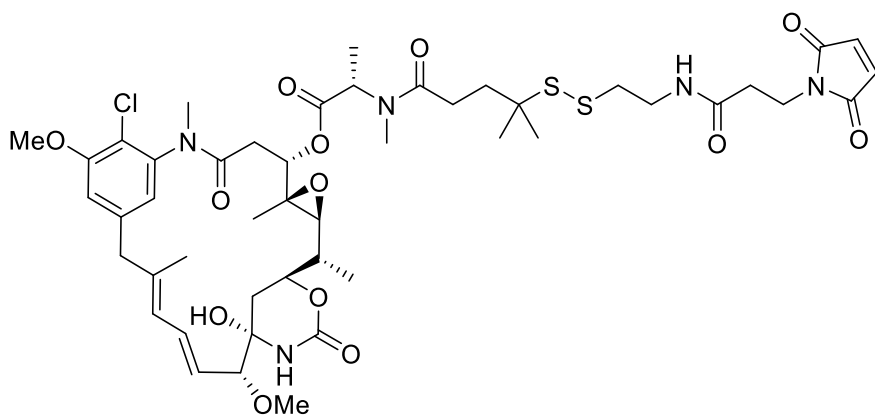
L² is a C₁₋₆alkylene or is -(CH₂CH₂O)_y-CH₂-CH₂- wherein y is 1 to 11;

X is -C(O)-NH-, -NHC(O)- or a triazole; and alkylene is linear or branched;

- (b) conjugating said linker-drug moiety to the antibody recovered from the cell culture of claim 46 to produce an antibody drug conjugate; and
- (c) purifying the antibody drug conjugate.

46. The process according to claim 45 comprising:

(a) pre-forming a linker-drug moiety of the following Formula:



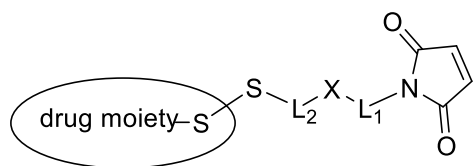
and

(b) conjugating said linker-drug moiety to the antibody recovered from the cell culture of claim 46 to produce an antibody drug conjugate; and

(c) purifying the antibody drug conjugate.

47. A process for producing an anti-CCR7 antibody drug conjugate comprising:

(a) pre-forming a linker-drug moiety of the following Formula:



wherein:

the drug moiety is DM1, DM3 or DM4 and the drug moiety is attached to the linker via its thiol functionality;

L^1 is a C_{1-6} alkylene wherein one of the methylene groups may be replaced with oxygen;

L^2 is a C_{1-6} alkylene or is $-(CH_2CH_2O)_y-CH_2-CH_2-$ wherein y is 1 to 11;

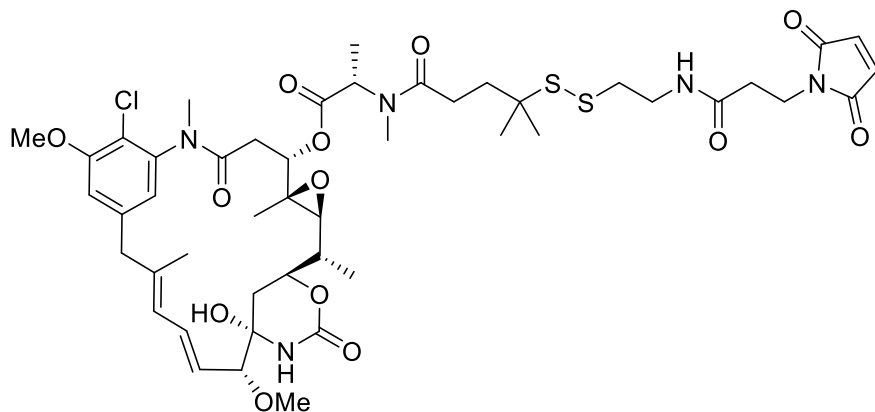
X is $-C(O)-NH-$, $-NHC(O)-$ or a triazole; and alkylene is linear or branched;

(b) conjugating said linker-drug moiety to the antibody of any one of claims 1-14 to produce an antibody drug conjugate; and

(c) purifying the antibody drug conjugate.

48. A process for producing an anti-CCR7 antibody drug conjugate comprising:

(a) pre-forming a linker-drug moiety of the following Formula:

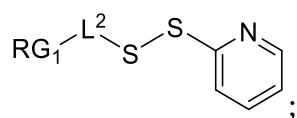


(b) conjugating said linker-drug moiety to the antibody of any one of claims 1-14 to produce an antibody drug conjugate; and

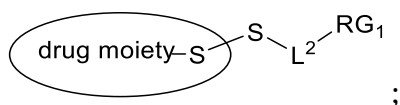
(c) purifying the antibody drug conjugate.

49. The process according to claim 45 or 47 wherein the step of pre-forming the linkerdrug moiety comprises:

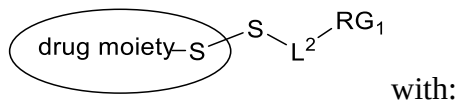
c) Reacting a drug moiety via its thiol functionality with:

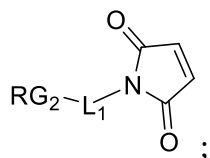


to form:

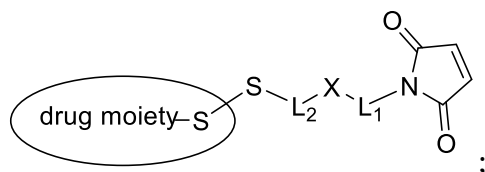


d) Reacting the formed





to form the linker-drug moiety:



wherein:

L^1 is a C_{1-6} alkylene wherein one of the methylene groups may be replaced with oxygen;

L^2 is a C_{1-6} alkylene or is $-(CH_2CH_2O)_y-CH_2-CH_2-$ wherein y is 1 to 11; and

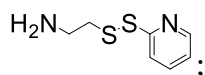
X is $-C(O)-NH-$, $-NHC(O)-$ or a triazole;

wherein the alkylene is linear or branched; and

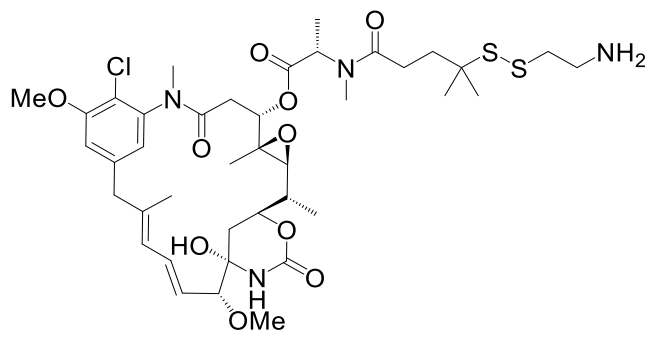
RG1 and RG2 are 2 reactive groups forming group X .

50. The process according to claim 46 or 48 wherein the step of pre-forming the linkerdrug moiety comprises:

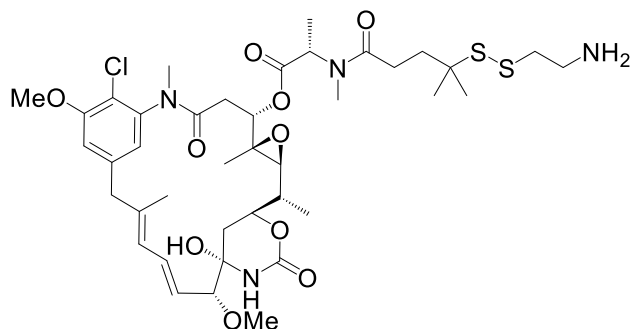
e) Reacting the drug moiety via its thiol functionality with:



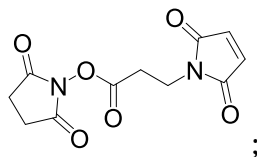
to form:



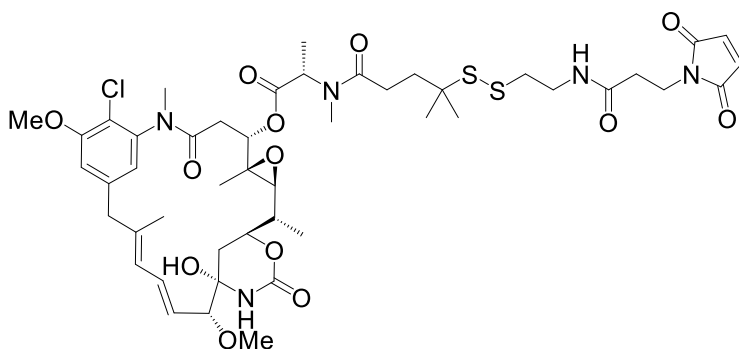
f) Reacting the formed



with:



to form the linker-drug moiety:



51. The antibody drug conjugate made according to any one of claims 45-50 having an average DAR, measured with a UV spectrophotometer, of about 3 to about 4.

52. A process for producing an anti-CCR7 antibody drug conjugate comprising:

- (a) chemically linking SMCC or MPET to a drug moiety DM-1 or DM-4 to form a linker-drug;
- (b) conjugating said linker-drug to the antibody as claimed in any one of claims 1-14; and
- (c) purifying the antibody drug conjugate.

53. The antibody drug conjugate made according to claim 52 having an average DAR, measured with a UV spectrophotometer, of about 3 to about 4.

54. A diagnostic reagent comprising the antibody or antigen binding fragment thereof of any one of claims 1-14.

55. The diagnostic reagent of claim 54, wherein the antibody or antigen binding fragment thereof is labeled with a radiolabel, a fluorophore, a chromophore, an imaging agent, or a metal ion.

56. The antibody or antigen binding fragment thereof of any one of claims 1-14, or the antibody drug conjugate of any one of claims 15-26, or the pharmaceutical composition of any one of claims 27 or 28 for use as a medicament.