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(54) Title: COMBINATION THERAPIES WITH ANTI-FOLATE RECEPTOR ANTIBODY CONJUGATES

(57) Abstract: The present disclosure relates to combination therapies with antibody conjugates with binding specificity for folate receptor alpha (FOLR1) and its isoforms and homologs, and compositions comprising combinations with the antibody conjugates, including pharmaceutical compositions. Also provided are methods of using the combinations with antibody conjugates and compositions thereof, such as in therapeutic and diagnostic methods.

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COMBINATION THERAPIES WITH ANTI-FOLATE RECEPTOR ANTIBODY CONJUGATES

FIELD OF THE INVENTION

[0001] Provided herein are combination therapies with antibody conjugates having binding specificity for folate receptor alpha (FolRα or FOLR1) and compositions for administering the same, including pharmaceutical compositions. The compositions are useful in methods of treatment and prevention of cell proliferation and cancer, methods of detection of cell proliferation and cancer, and methods of diagnosis of cell proliferation and cancer. The compositions are also useful in methods of treatment, prevention, detection, and diagnosis of autoimmune diseases, infectious diseases, and inflammatory conditions.

BACKGROUND

[0002] Folate receptors, or folate binding proteins (FBPs), include single chain glycoproteins that bind and contribute to the update of folates and other compounds *in vivo*. Elwood, 1989, *J. Biol. Chem.* 264:14893-14901. Certain folate receptors are single-chain glycoproteins with a high affinity binding site for folate and other compounds such as methotrexate. Elwood, p. 14893. The human FOLR1 gene encodes the adult folate receptor, a 30 kDa polypeptide with about 257 amino acids with three potential N-linked glycosylation sites. Elwood, p. 14893; Lacey *et al.*, 1989, *J. Clin. Invest.* 84:715-720. Homologous genes and polypeptides have been identified in dozens of species.

[0003] The mature folate receptor glycoprotein has a size of about 42 kDa and has been observed to participate in the internalization of folates and antifolates into cells. Elwood *et al.*, 1997, *Biochemistry* 36:1467-1478. Expression has been observed in human cerebellum and kidney cells, along with human cancer cell lines. Elwood *et al.*, 1997, p. 1467. In addition to internalization of folate, a folate receptor has been shown to be a significant cofactor for cellular entry of viruses, particularly Marburg and Ebola viruses. Chan *et al.*, 2001, *Cell* 106:117-126. Due to these internalization properties, the folate receptor has been proposed as a target for diagnostic and therapeutic agents. For instance, diagnostic and therapeutic agents have been linked to folate for internalization into cells expressing the folate receptor. *See, e.g.,* Leamon, 2008, *Curr. Opin. Investig. Drugs* 9:1277-1286; Paulos *et al.*, 2004, *Adv. Drug Del. Rev.* 56:1205-1217.

[0004] Folate receptor alpha (FolRα or FOLR1) is a glycosylphosphatidylinositol linked cell-surface glycoprotein that has high affinity for folates. Except for low levels in kidney and lung, most normal tissues do not express FOLR1, but high levels of FOLR1 have been found in serous and endometrioid epithelial ovarian cancer, endometrial adenocarcinoma, non-small cell lung carcinoma (NSCLC) of the adenocarcinoma subtype, and triple-negative breast cancer (TNBC). FOLR1 expression is maintained in metastatic foci and recurrent carcinomas in ovarian cancer patients, and FOLR1 expression has been observed after chemotherapy in epithelial ovarian and endometrial cancers. These properties, together with the highly restricted expression of FOLR1 on normal tissues, make FOLR1 a highly promising target for cancer therapy. As such, the folate receptor provides a potential target for diagnostics and therapeutics for cancers and inflammatory conditions. In addition to a need for new antibodies for specific binding and targeting of these folate receptors, combination therapies with other anti-cancer agents or agents that target cell division and/or cell differentiation provide another avenue of exploration for treatment of diseases and disorders associated with overexpression FOLR1 and/or hyperactivity of FOLR1 signaling.

[0005] There is a need for improved methods of modulating the immune regulation of folate receptor alpha (FOLR1) and the downstream signaling processes activated by folate receptor alpha (FOLR1), which may be enhanced or improved in combination with administered anti-cancer agents or therapeutic agents that modulate cell division and/or cell differentiation. Such therapeutic agents, in combination with anti-FOLR1 antibody conjugates that deliver therapeutic or diagnostic payload moieties to target cells expressing folate receptor alpha, may be useful in the treatment or diagnosis of diseases in which FOLR1 is overexpressed.

SUMMARY

[0006] Provided herein are antibody conjugates that selectively bind folate receptor alpha (FOLR1), which are administered in combination with a second therapeutic agent that modulates cell division or cell differentiation activity. The antibody conjugates comprise an antibody that binds folate receptor alpha (FOLR1) linked to one or more payload moieties. The antibody can be linked to the payload directly by a covalent bond or indirectly by way of a linker. Folate receptor alpha (FOLR1) antibodies are described in detail herein, as are useful payload moieties, and useful linkers. In a particular embodiment, the second therapeutic agent is an inhibitor of programmed cell death protein (PD-1).

[0007] In another aspect, provided are compositions comprising the antibody conjugates and a second therapeutic agent that modulates cell division or cell differentiation activity. In some embodiments, the compositions are pharmaceutical compositions. Any suitable pharmaceutical composition may be used. In some embodiments, the pharmaceutical composition is a composition for parenteral administration. In a further aspect, provided herein are kits comprising the antibody conjugates or pharmaceutical compositions. In a particular embodiment, the second therapeutic agent is an inhibitor of programmed cell death protein (PD-1).

[0008] In another aspect, provide herein are methods of using the anti-FOLR1 antibody conjugates in combination with a second therapeutic agent that modulates cell division or cell differentiation activity. In some embodiments, the methods are methods of delivering the combination to a target cell or tissue expressing folate receptor alpha. In some embodiments, the methods are methods of treatment. In some embodiments, the methods are diagnostic methods. In some embodiments, the methods are analytical methods. In some embodiments, the combination is used to treat a disease or condition. In some aspects, the disease or condition is selected from a cancer, autoimmune disease, and infection.

[0009] In some embodiments, the antibody conjugates bind human folate receptor alpha. In some embodiments, the antibody conjugates also bind homologs of human folate receptor alpha. In some aspects, the antibody conjugates also bind homologs of cynomolgus monkey and/or mouse folate receptor alpha.

BRIEF DESCRIPTION OF THE FIGURES

[0010] **FIG. 1** provides a comparison of the Kabat and Chothia numbering systems for CDR-H1. *Adapted from* Martin A.C.R. (2010). Protein Sequence and Structure Analysis of Antibody Variable Domains. In R. Kontermann & S. Dübel (Eds.), *Antibody Engineering* vol. 2 (pp. 33-51). Springer-Verlag, Berlin Heidelberg.

[0011] **FIGS. 2-4** provide alignments of the VH sequences (SEQ ID NOs: 308-366) from the variant antibodies provided herein. CDRs according to Chothia are highlighted, and CDRs according to Kabat are boxed.

[0012] **FIG. 5** provides alignments of the VL sequences (SEQ ID NOs: 367-369) from trastuzumab and the variant antibodies provided herein. CDRs according to Chothia are highlighted, and CDRs according to Kabat are underlined.

[0013] **FIG. 6** is a graph illustrating body weight change in mice implanted with KB cervical carcinoma cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0014] **FIG. 7 (A, B)** are graphs illustrating tumor growth curves and tumor size at day 25 in mice implanted with KB cervical carcinoma cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0015] **FIG. 8** is a scatter plot illustrating final tumor size at day 31 in mice implanted with KB cervical carcinoma cells after single-dose treatment with two different FOLR1 antibody-drug conjugates disclosed herein.

[0016] **FIG. 9** is a graph illustrating body weight change in mice implanted with KB cervical carcinoma cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0017] **FIG. 10 (A, B)** are graphs illustrating tumor growth curves and tumor size at day 21 in mice implanted with KB cervical carcinoma cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0018] **FIG. 11** is a scatter plot illustrating final tumor size at day 36 in mice implanted with KB cervical carcinoma cells after single-dose treatment with three different FOLR1 antibody-drug conjugates disclosed herein.

[0019] **FIG. 12** is a graph illustrating body weight change in mice implanted with Igrov1 ovarian cancer cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0020] **FIG. 13 (A, B)** are graphs illustrating tumor growth curves and tumor size at day 24 in mice implanted with Igrov1 ovarian cancer cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0021] **FIG. 14A** is a graph illustrating tumor growth curves in mice implanted with Igrov1 ovarian cancer cells after being administered a single dose of different FOLR1 antibody-drug conjugates as disclosed herein.

[0022] **FIG. 14B** is a scatter plot illustrating tumor size at day 21 in mice implanted with Igrov1 ovarian cancer cells after single-dose treatment with different FOLR1 antibody-drug conjugates disclosed herein.

[0023] **FIG. 15** includes plots illustrating binding of different FOLR1 antibodies to 293T transformed cells stably expressing different folate receptor isoforms (hFOLR1, hFOLR2).

[0024] **FIG. 16** includes plots illustrating the cytotoxic activity of different FOLR1 antibody-drug conjugates on 293T transformed cells stably expressing different folate receptor isoforms (hFOLR1, hFOLR2).

[0025] **FIG. 17** is a graph illustrating body weight change in mice implanted with Igrov1 ovarian cancer cells after being administered various doses of different FOLR1 antibody-drug conjugates as disclosed herein.

[0026] **FIG. 18 (A, B, C)** includes tumor growth curves and scatter plot with tumor size at day 21 in mice implanted with Igrov1 ovarian cancer cells after being administered various doses of different FOLR1 antibody-drug conjugates as disclosed herein.

[0027] **FIG. 19 (A, B, C, D)** includes graphs illustrating tumor size in mice implanted with Igrov1 ovarian cancer cells after being administered various doses of different FOLR1 antibody-drug conjugates as disclosed herein.

[0028] **FIG. 20** is a chart illustrating the delay of tumor growth in mice implanted with Igrov1 ovarian cancer cells after being administered various doses of different FOLR1 antibody-drug conjugates as disclosed herein.

[0029] **FIG. 21 (A, B, C)** includes tumor growth charts, a scatter plot illustrating tumor size at day 29, and a chart illustrating tumor growth inhibition at day 29 in animals bearing established Igrov1 tumors treated with a single dose of an exemplary FOLR-1 antibody-drug conjugate with or without carboplatin.

[0030] **FIG. 22 (A, B)** includes a tumor growth chart and a scatter plot illustrating tumor size at day 31 in animals bearing established OVCAR3 tumors.

[0031] **FIG. 23 (A-G)** includes tumor growth curves of various endometrium patient derived xenograft models to which an exemplary FOLR1 antibody-drug conjugate was administered.

[0032] **FIG. 24 (A, B)** includes tumor growth curves and a tumor size scatter plot of animals with MC38-hFOLR1 tumors in response to treatment with an exemplary FOLR1 antibody-drug conjugate, Avelumab, or a combination of both.

[0033] **FIG. 25 (A, B)** includes tumor growth curves and a Kaplan-Meier survival plot of animals with MC38-hFOLR1 tumors in response to treatment with an exemplary FOLR1 antibody-drug conjugate, Avelumab, or a combination of both.

[0034] **FIG. 26** is a graph illustrating the pharmacokinetic plasma profile of different FOLR1 antibody-drug conjugates in SCID Beige mice.

[0035] **FIG. 27** is an LC/MS trace of small molecules detected in the plasma of mice treated with vehicle or with ADC Molecules 1 or 17.

[0036] **FIG. 28** includes a graph illustrating the plasma stability (as measured by drug-antibody ratio, or DAR) of a representative FOLR1 antibody-drug conjugate administered to SCID Beige mice.

[0037] **FIG. 29** includes graphs illustrating the plasma stability (as measured by drug-antibody ratio, or DAR) of various FOLR1 antibody-drug conjugates as tested in PBS, cynomolgous monkey plasma, or human plasma.

[0038] **FIG. 30** includes graphs illustrating the cytotoxic activity of ADC Molecule 4 and ADC Molecule 21 on various cells in the presence of the respective naked antibody as competitor.

[0039] **FIG. 31** is a graph illustrating the body weight change in rats that were administered various doses of the catabolites of FOLR1 antibody-drug conjugates disclosed herein.

[0040] **FIG. 32** is a graph illustrating the stability of a representative FOLR1 antibody-drug conjugate (ADC) compared to a comparator ADC as tested in cynomolgous monkey plasma, human plasma, and PBS.

[0041] **FIG. 33** includes graphs illustrating cytotoxic activity of the catabolites of a representative FOLR1 antibody-drug conjugate (ADC) disclosed herein compared to that of a comparator ADC in cells with varying levels of PgP and in the presence of a specific PgP inhibitor.

[0042] **FIG. 34** is a chart illustrating tumor and plasma levels of the catabolite of a representative FOLR1 antibody-drug conjugate (ADC) disclosed herein compared to that of a comparator ADC as measured in mice with established Igrov1 tumors.

[0043] **FIG. 35 (A, B)** includes a tumor growth curve and a scatter plot illustrating tumor size at day 21 for different FOLR1 antibody-drug conjugates (ADC) as disclosed herein.

[0044] **FIG. 36** is a graph illustrating the pharmacokinetic plasma profile of different FOLR1 antibody-drug conjugates in SCID Beige mice.

DETAILED DESCRIPTION OF THE EMBODIMENTS

1. Definitions

[0045] Unless otherwise defined, all terms of art, notations and other scientific terminology used herein are intended to have the meanings commonly understood by those of skill in the art to which this invention pertains. In some cases, terms with commonly understood meanings are defined herein for clarity and/or for ready reference, and the inclusion of such definitions herein should not necessarily be construed to represent a difference over what is generally understood in the art. The techniques and procedures described or referenced herein are generally well understood and commonly employed using conventional methodologies by those skilled in the art, such as, for example, the widely utilized molecular cloning methodologies described in Sambrook et al., *Molecular Cloning: A Laboratory Manual* 2nd ed. (1989) Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY. As appropriate, procedures involving the use of commercially available kits and reagents are generally carried out in accordance with manufacturer-defined protocols and conditions unless otherwise noted.

[0046] As used herein, the singular forms "a," "an," and "the" include the plural referents unless the context clearly indicates otherwise.

[0047] The term "about" indicates and encompasses an indicated value and a range above and below that value. In certain embodiments, the term "about" indicates the designated value ± 10%, ± 5%, or ± 1%. In certain embodiments, the term "about" indicates the designated value ± one standard deviation of that value.

[0048] The term "combinations thereof" includes every possible combination of elements to which the term refers to. For example, a sentence stating that "if α_2 is A, then α_3 is not D; α_5 is not S; or α_6 is not S; *or combinations thereof*" includes the following combinations when α_2 is A: (1) α_3 is not D; (2) α_5 is not S; (3) α_6 is not S; (4) α_3 is not D; α_5 is not S; *and* α_6 is not S; (5) α_3 is not D *and* α_5 is not S; (6) α_3 is not D *and* α_6 is not S; and (7) α_5 is not S *and* α_6 is not S.

[0049] The terms "folate receptor alpha" and "folate receptor 1" are used interchangeably herein. Folate receptor alpha is also known by synonyms, including FOLR1, FolRα, folate binding protein, FBP, adult folate binding protein, Folbp1, FR-alpha, FRα, KB cells FBP, and

ovarian tumor-associated antigen MOv18, among others. Unless specified otherwise, the terms include any variants, isoforms and species homologs of human folate receptor alpha that are naturally expressed by cells, or that are expressed by cells transfected with a folate receptor alpha or *FOLR1* gene. Folate receptor alpha proteins include, for example, human folate receptor alpha (SEQ ID NO: 1). In some embodiments, folate receptor alpha proteins include cynomolgus monkey folate receptor alpha (SEQ ID NO: 2). In some embodiments, folate receptor alpha proteins include murine folate receptor alpha (SEQ ID NO: 3).

[0050] The term "immunoglobulin" refers to a class of structurally related proteins generally comprising two pairs of polypeptide chains: one pair of light (L) chains and one pair of heavy (H) chains. In an "intact immunoglobulin," all four of these chains are interconnected by disulfide bonds. The structure of immunoglobulins has been well characterized. *See, e.g.,* Paul, *Fundamental Immunology* 7th ed., Ch. 5 (2013) Lippincott Williams & Wilkins, Philadelphia, PA. Briefly, each heavy chain typically comprises a heavy chain variable region (V_H) and a heavy chain constant region (C_H). The heavy chain constant region typically comprises three domains, abbreviated C_{H1}, C_{H2}, and C_{H3}. Each light chain typically comprises a light chain variable region (V_L) and a light chain constant region. The light chain constant region typically comprises one domain, abbreviated C_L.

[0051] The term "antibody" describes a type of immunoglobulin molecule and is used herein in its broadest sense. An antibody specifically includes intact antibodies (e.g., intact immunoglobulins), and antibody fragments. Antibodies comprise at least one antigen-binding domain. One example of an antigen-binding domain is an antigen binding domain formed by a V_H -V_L dimer. A "folate receptor alpha antibody," "anti-folate receptor alpha antibody," "folate receptor alpha Ab," "folate receptor alpha-specific antibody," "anti-folate receptor alpha Ab," "FOLR1 antibody," "FolRα antibody," "anti-FOLR1 antibody," "anti-FolRα antibody," "FOLR1 Ab," "FolRα Ab," "FOLR1-specific antibody," "FolRα-specific antibody," "anti-FolRα Ab," or "anti-FOLR1 Ab" is an antibody, as described herein, which binds specifically to folate receptor alpha or FOLR1. In some embodiments, the antibody binds the extracellular domain of folate receptor alpha (FOLR1).

[0052] The V_H and V_L regions may be further subdivided into regions of hypervariability ("hypervariable regions (HVRs);" also called "complementarity determining regions" (CDRs)) interspersed with regions that are more conserved. The more conserved regions are called framework regions (FRs). Each V_H and V_L generally comprises three CDRs and four FRs,

arranged in the following order (from N-terminus to C-terminus): FR1 - CDR1 - FR2 - CDR2 - FR3 - CDR3 - FR4. The CDRs are involved in antigen binding, and influence antigen specificity and binding affinity of the antibody. See Kabat et al., *Sequences of Proteins of Immunological Interest* 5th ed. (1991) Public Health Service, National Institutes of Health, Bethesda, MD, incorporated by reference in its entirety.

[0053] The light chain from any vertebrate species can be assigned to one of two types, called kappa and lambda, based on the sequence of the constant domain.

[0054] The heavy chain from any vertebrate species can be assigned to one of five different classes (or isotypes): IgA, IgD, IgE, IgG, and IgM. These classes are also designated α, δ, ε, γ, and μ, respectively. The IgG and IgA classes are further divided into subclasses on the basis of differences in sequence and function. Humans express the following subclasses: IgG1, IgG2, IgG3, IgG4, IgA1, and IgA2.

[0055] The amino acid sequence boundaries of a CDR can be determined by one of skill in the art using any of a number of known numbering schemes, including those described by Kabat et al., *supra* ("Kabat" numbering scheme); Al-Lazikani et al., 1997, *J. Mol. Biol.*, 273:927-948 ("Chothia" numbering scheme); MacCallum et al., 1996, *J. Mol. Biol.* 262:732-745 ("Contact" numbering scheme); Lefranc et al., *Dev. Comp. Immunol.*, 2003, 27:55-77 ("IMGT" numbering scheme); and Honegge and Plückthun, *J. Mol. Biol.*, 2001, 309:657-70 ("AHo" numbering scheme), each of which is incorporated by reference in its entirety.

[0056] Table 1 provides the positions of CDR-L1, CDR-L2, CDR-L3, CDR-H1, CDR-H2, and CDR-H3 as identified by the Kabat and Chothia schemes. For CDR-H1, residue numbering is provided using both the Kabat and Chothia numbering schemes.

Table 1. Residues in CDRs according to Kabat and Chothia numbering schemes.

CDR	Kabat	Chothia
L1	L24-L34	L24-L34
L2	L50-L56	L50-L56
L3	L89-L97	L89-L97
H1 (Kabat Numbering)	H31-H35B	H26-H32 or H34*
H1 (Chothia Numbering)	H31-H35	H26-H32
H2	H50-H65	H52-H56
H3	H95-H102	H95-H102

* The C-terminus of CDR-H1, when numbered using the Kabat numbering convention, varies between H32 and H34, depending on the length of the CDR, as illustrated in FIG. 1.

[0057] Unless otherwise specified, the numbering scheme used for identification of a particular CDR herein is the Kabat/Chothia numbering scheme. Where the residues encompassed by these two numbering schemes diverge (e.g., CDR-H1 and/or CDR-H2), the numbering scheme is specified as either Kabat or Chothia. For convenience, CDR-H3 is sometimes referred to herein as either Kabat or Chothia. However, this is not intended to imply differences in sequence where they do not exist, and one of skill in the art can readily confirm whether the sequences are the same or different by examining the sequences.

[0058] CDRs may be assigned, for example, using antibody numbering software, such as Abnum, available at www.bioinf.org.uk/abs/abnum/, and described in Abhinandan and Martin, *Immunology*, 2008, 45:3832-3839, incorporated by reference in its entirety.

[0059] The "EU numbering scheme" is generally used when referring to a residue in an antibody heavy chain constant region (e.g., as reported in Kabat et al., *supra*). Unless stated otherwise, the EU numbering scheme is used to refer to residues in antibody heavy chain constant regions described herein.

[0060] An "antibody fragment" comprises a portion of an intact antibody, such as the antigen binding or variable region of an intact antibody. Antibody fragments include, for example, Fv fragments, Fab fragments, $F(ab')_2$ fragments, Fab' fragments, scFv (sFv) fragments, and scFv-Fc fragments.

[0061] "Fv" fragments comprise a non-covalently-linked dimer of one heavy chain variable domain and one light chain variable domain.

[0062] "Fab" fragments comprise, in addition to the heavy and light chain variable domains, the constant domain of the light chain and the first constant domain (C_{H1}) of the heavy chain. Fab fragments may be generated, for example, by recombinant methods or by papain digestion of a full-length antibody.

[0063] "$F(ab')_2$" fragments contain two Fab' fragments joined, near the hinge region, by disulfide bonds. $F(ab')_2$ fragments may be generated, for example, by recombinant methods or by pepsin digestion of an intact antibody. The F(ab') fragments can be dissociated, for example, by treatment with ß-mercaptoethanol.

[0064] "Single-chain Fv" or "sFv" or "scFv" antibody fragments comprise a V_H domain and a V_L domain in a single polypeptide chain. The V_H and V_L are generally linked by a peptide linker. *See* Plückthun A. (1994). In some embodiments, the linker is SEQ ID NO: 377. In some

embodiments, the linker is SEQ ID NO: 378. Antibodies from *Escherichia coli*. In Rosenberg M. & Moore G.P. (Eds.), *The Pharmacology of Monoclonal Antibodies* vol. 113 (pp. 269-315). Springer-Verlag, New York, incorporated by reference in its entirety.

[0065] "scFv-Fc" fragments comprise an scFv attached to an Fc domain. For example, an Fc domain may be attached to the C-terminus of the scFv. The Fc domain may follow the V_H or V_L, depending on the orientation of the variable domains in the scFv (i.e., V_H -V_L or V_L -V_H). Any suitable Fc domain known in the art or described herein may be used. In some cases, the Fc domain comprises an IgG1 Fc domain. In some embodiments, the IgG1 Fc domain comprises SEQ ID NO: 370, or a portion thereof. SEQ ID NO: 370 provides the sequence of C_{H1}, C_{H2}, and C_{H3} of the human IgG1 constant region.

[0066] The term "monoclonal antibody" refers to an antibody from a population of substantially homogeneous antibodies. A population of substantially homogeneous antibodies comprises antibodies that are substantially similar and that bind the same epitope(s), except for variants that may normally arise during production of the monoclonal antibody. Such variants are generally present in only minor amounts. A monoclonal antibody is typically obtained by a process that includes the selection of a single antibody from a plurality of antibodies. For example, the selection process can be the selection of a unique clone from a plurality of clones, such as a pool of hybridoma clones, phage clones, yeast clones, bacterial clones, or other recombinant DNA clones. The selected antibody can be further altered, for example, to improve affinity for the target ("affinity maturation"), to humanize the antibody, to improve its production in cell culture, and/or to reduce its immunogenicity in a subject.

[0067] The term "chimeric antibody" refers to an antibody in which a portion of the heavy and/or light chain is derived from a particular source or species, while the remainder of the heavy and/or light chain is derived from a different source or species.

[0068] "Humanized" forms of non-human antibodies are chimeric antibodies that contain minimal sequence derived from the non-human antibody. A humanized antibody is generally a human immunoglobulin (recipient antibody) in which residues from one or more CDRs are replaced by residues from one or more CDRs of a non-human antibody (donor antibody). The donor antibody can be any suitable non-human antibody, such as a mouse, rat, rabbit, chicken, or non-human primate antibody having a desired specificity, affinity, or biological effect. In some instances, selected framework region residues of the recipient antibody are replaced by the corresponding framework region residues from the donor antibody. Humanized antibodies

may also comprise residues that are not found in either the recipient antibody or the donor antibody. Such modifications may be made to further refine antibody function. For further details, *see* Jones et al., *Nature*, 1986, 321:522-525; Riechmann et al., *Nature*, 1988, 332:323-329; and Presta, *Curr. Op. Struct. Biol.*, 1992, 2:593-596, each of which is incorporated by reference in its entirety.

[0069] A "human antibody" is one which possesses an amino acid sequence corresponding to that of an antibody produced by a human or a human cell, or derived from a non-human source that utilizes a human antibody repertoire or human antibody-encoding sequences (e.g., obtained from human sources or designed *de novo*). Human antibodies specifically exclude humanized antibodies.

[0070] An "isolated antibody" is one that has been separated and/or recovered from a component of its natural environment. Components of the natural environment may include enzymes, hormones, and other proteinaceous or nonproteinaceous materials. In some embodiments, an isolated antibody is purified to a degree sufficient to obtain at least 15 residues of N-terminal or internal amino acid sequence, for example by use of a spinning cup sequenator. In some embodiments, an isolated antibody is purified to homogeneity by gel electrophoresis (e.g., SDS-PAGE) under reducing or nonreducing conditions, with detection by Coomassie blue or silver stain. An isolated antibody includes an antibody *in situ* within recombinant cells, since at least one component of the antibody's natural environment is not present. In some aspects, an isolated antibody is prepared by at least one purification step.

[0071] In some embodiments, an isolated antibody is purified to at least 80%, 85%, 90%, 95%, or 99% by weight. In some embodiments, an isolated antibody is purified to at least 80%, 85%, 90%, 95%, or 99% by volume. In some embodiments, an isolated antibody is provided as a solution comprising at least 85%, 90%, 95%, 98%, 99% to 100% by weight. In some embodiments, an isolated antibody is provided as a solution comprising at least 85%, 90%, 95%, 98%, 99% to 100% by volume.

[0072] "Affinity" refers to the strength of the sum total of non-covalent interactions between a single binding site of a molecule (e.g., an antibody) and its binding partner (e.g., an antigen). Unless indicated otherwise, as used herein, "binding affinity" refers to intrinsic binding affinity, which reflects a 1:1 interaction between members of a binding pair (e.g., antibody and antigen). The affinity of a molecule X for its partner Y can be represented by the dissociation constant (K_D). Affinity can be measured by common methods known in the art,

including those described herein. Affinity can be determined, for example, using surface plasmon resonance (SPR) technology, such as a Biacore® instrument. In some embodiments, the affinity is determined at 25°C.

[0073] With regard to the binding of an antibody to a target molecule, the terms "specific binding," "specifically binds to," "specific for," "selectively binds," and "selective for" a particular antigen (e.g., a polypeptide target) or an epitope on a particular antigen mean binding that is measurably different from a non-specific or non-selective interaction. Specific binding can be measured, for example, by determining binding of a molecule compared to binding of a control molecule. Specific binding can also be determined by competition with a control molecule that mimics the antibody binding site on the target. In that case, specific binding is indicated if the binding of the antibody to the target is competitively inhibited by the control molecule.

[0074] The term "k_d" (sec^{-1}), as used herein, refers to the dissociation rate constant of a particular antibody-antigen interaction. This value is also referred to as the k_{off} value.

[0075] The term "k_a" ($M^{-1} \times sec^{-1}$), as used herein, refers to the association rate constant of a particular antibody-antigen interaction. This value is also referred to as the k_{on} value.

[0076] The term "K_D" (M), as used herein, refers to the dissociation equilibrium constant of a particular antibody-antigen interaction. $K_D = k_d/k_a$.

[0077] The term "K_A" (M^{-1}), as used herein, refers to the association equilibrium constant of a particular antibody-antigen interaction. $K_A = k_a/k_d$.

[0078] An "affinity matured" antibody is one with one or more alterations in one or more CDRs or FRs that result in an improvement in the affinity of the antibody for its antigen, compared to a parent antibody which does not possess the alteration(s). In one embodiment, an affinity matured antibody has nanomolar or picomolar affinity for the target antigen. Affinity matured antibodies may be produced using a variety of methods known in the art. For example, Marks et al. (*Bio/Technology*, 1992, 10:779-783, incorporated by reference in its entirety) describes affinity maturation by V_H and V_L domain shuffling. Random mutagenesis of CDR and/or framework residues is described by, for example, Barbas et al. (*Proc. Nat. Acad. Sci. U.S.A.*, 1994, 91:3809-3813); Schier et al., *Gene*, 1995, 169:147-155; Yelton et al., *J. Immunol.*, 1995, 155:1994-2004; Jackson et al., *J. Immunol.*, 1995, 154:3310-33199; and Hawkins et al, *J. Mol. Biol.*, 1992, 226:889-896, each of which is incorporated by reference in its entirety.

[0079] When used herein in the context of two or more antibodies, the term "competes with" or "cross-competes with" indicates that the two or more antibodies compete for binding to an antigen (e.g., folate receptor alpha, or FOLR1). In one exemplary assay, FOLR1 is coated on a plate and allowed to bind a first antibody, after which a second, labeled antibody is added. If the presence of the first antibody reduces binding of the second antibody, then the antibodies compete. In another exemplary assay, a first antibody is coated on a plate and allowed to bind the antigen, and then the second antibody is added. The term "competes with" also includes combinations of antibodies where one antibody reduces binding of another antibody, but where no competition is observed when the antibodies are added in the reverse order. However, in some embodiments, the first and second antibodies inhibit binding of each other, regardless of the order in which they are added. In some embodiments, one antibody reduces binding of another antibody to its antigen by at least 50%, at least 60%, at least 70%, at least 80%, or at least 90%.

[0080] The term "epitope" means a portion of an antigen capable of specific binding to an antibody. Epitopes frequently consist of surface-accessible amino acid residues and/or sugar side chains and may have specific three dimensional structural characteristics, as well as specific charge characteristics. Conformational and non-conformational epitopes are distinguished in that the binding to the former but not the latter is lost in the presence of denaturing solvents. An epitope may comprise amino acid residues that are directly involved in the binding, and other amino acid residues, which are not directly involved in the binding. The epitope to which an antibody binds can be determined using known techniques for epitope determination such as, for example, testing for antibody binding to variants of folate receptor alpha (FOLR1) with different point-mutations.

[0081] Percent "identity" between a polypeptide sequence and a reference sequence, is defined as the percentage of amino acid residues in the polypeptide sequence that are identical to the amino acid residues in the reference sequence, after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent sequence identity. Alignment for purposes of determining percent amino acid sequence identity can be achieved in various ways that are within the skill in the art, for instance, using publicly available computer software such as BLAST, BLAST-2, ALIGN, MEGALIGN (DNASTAR), CLUSTALW, CLUSTAL OMEGA, or MUSCLE software. Those skilled in the art can determine appropriate parameters for aligning sequences, including any algorithms needed to achieve maximal alignment over the full length of the sequences being compared.

[0082] A "conservative substitution" or a "conservative amino acid substitution," refers to the substitution of an amino acid with a chemically or functionally similar amino acid. Conservative substitution tables providing similar amino acids are well known in the art. Polypeptide sequences having such substitutions are known as "conservatively modified variants." Such conservatively modified variants are in addition to and do not exclude polymorphic variants, interspecies homologs, and alleles. By way of example, the groups of amino acids provided in Tables 2-4 are, in some embodiments, considered conservative substitutions for one another.

Table 2. Selected groups of amino acids that are considered conservative substitutions for one another, in certain embodiments.

Acidic Residues	D and E
Basic Residues	K, R, and H
Hydrophilic Uncharged Residues	S, T, N, and Q
Aliphatic Uncharged Residues	G, A, V, L, and I
Non-polar Uncharged Residues	C, M, and P
Aromatic Residues	F, Y, and W
Alcohol Group-Containing Residues	S and T
Aliphatic Residues	I, L, V, and M
Cycloalkenyl-associated Residues	F, H, W, and Y
Hydrophobic Residues	A, C, F, G, H, I, L, M, R, T, V, W, and Y
Negatively Charged Residues	D and E
Polar Residues	C, D, E, H, K, N, Q, R, S, and T
Positively Charged Residues	H, K, and R
Small Residues	A, C, D, G, N, P, S, T, and V
Very Small Residues	A, G, and S
Residues Involved in Turn Formation	A, C, D, E, G, H, K, N, Q, R, S, P, and T
Flexible Residues	Q, T, K, S, G, P, D, E, and R

Table 3. Additional selected groups of amino acids that are considered conservative substitutions for one another, in certain embodiments.

Group 1	A, S, and T
Group 2	D and E
Group 3	N and Q
Group 4	R and K
Group 5	I, L, and M
Group 6	F, Y, and W

Table 4. Further selected groups of amino acids that are considered conservative substitutions for one another, in certain embodiments.

Group A	A and G
Group B	D and E
Group C	N and Q
Group D	R, K, and H
Group E	I, L, M, V
Group F	F, Y, and W
Group G	S and T
Group H	C and M

[0083] Additional conservative substitutions may be found, for example, in Creighton, *Proteins: Structures and Molecular Properties* 2nd ed. (1993) W. H. Freeman & Co., New York, NY. An antibody generated by making one or more conservative substitutions of amino acid residues in a parent antibody is referred to as a "conservatively modified variant."

[0084] The term "amino acid" refers to the twenty common naturally occurring amino acids. Naturally occurring amino acids include alanine (Ala; A), arginine (Arg; R), asparagine (Asn; N), aspartic acid (Asp; D), cysteine (Cys; C); glutamic acid (Glu; E), glutamine (Gln; Q), Glycine (Gly; G); histidine (His; H), isoleucine (Ile; I), leucine (Leu; L), lysine (Lys; K), methionine (Met; M), phenylalanine (Phe; F), proline (Pro; P), serine (Ser; S), threonine (Thr; T), tryptophan (Trp; W), tyrosine (Tyr; Y), and valine (Val; V).

[0085] Naturally encoded amino acids are the proteinogenic amino acids known to those of skill in the art. They include the 20 common amino acids (alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, and valine) and the less common pyrrolysine and selenocysteine. Naturally encoded amino acids include post-translational variants of the 22 naturally occurring amino acids such as prenylated amino acids, isoprenylated amino acids, myrisoylated amino acids, palmitoylated amino acids, N-linked glycosylated amino acids, O-linked glycosylated amino acids, phosphorylated amino acids and acylated amino acids.

[0086] The term "non-natural amino acid" refers to an amino acid that is not a proteinogenic amino acid, or a post-translationally modified variant thereof. In particular, the term refers to an amino acid that is not one of the 20 common amino acids or pyrrolysine or selenocysteine, or post-translationally modified variants thereof.

[0087] The term "conjugate" or "antibody conjugate" refers to an antibody linked to one or more payload moieties. The antibody can be any antibody described herein. The payload

can be any payload described herein. The antibody can be directly linked to the payload via a covalent bond, or the antibody can be linked to the payload indirectly via a linker. Typically, the linker is covalently bonded to the antibody and also covalently bonded to the payload. The term "antibody drug conjugate" or "ADC" refers to a conjugate wherein at least one payload is a therapeutic moiety such as a drug.

[0088] The term "payload" refers to a molecular moiety that can be conjugated to an antibody. In particular embodiments, payloads are selected from the group consisting of therapeutic moieties and labelling moieties.

[0089] The term "linker" refers to a molecular moiety that is capable of forming at least two covalent bonds. Typically, a linker is capable of forming at least one covalent bond to an antibody and at least another covalent bond to a payload. In certain embodiments, a linker can form more than one covalent bond to an antibody. In certain embodiments, a linker can form more than one covalent bond to a payload or can form covalent bonds to more than one payload. After a linker forms a bond to an antibody, or a payload, or both, the remaining structure, *i.e.* the residue of the linker after one or more covalent bonds are formed, may still be referred to as a "linker" herein. The term "linker precursor" refers to a linker having one or more reactive groups capable of forming a covalent bond with an antibody or payload, or both. In some embodiments, the linker is a cleavable linker. For example, a cleavable linker can be one that is released by an bio-labile function, which may or may not be engineered. In some embodiments, the linker is a non-cleavable linker. For example, a non-cleavable linker can be one that is released upon degradation of the antibody.

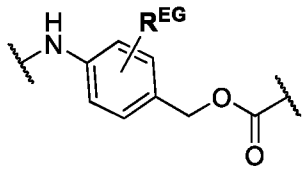
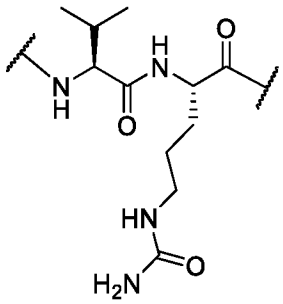
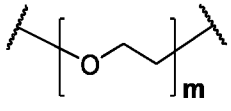
[0090] "Treating" or "treatment" of any disease or disorder refers, in certain embodiments, to ameliorating a disease or disorder that exists in a subject. In another embodiment, "treating" or "treatment" includes ameliorating at least one physical parameter, which may be indiscernible by the subject. In yet another embodiment, "treating" or "treatment" includes modulating the disease or disorder, either physically (e.g., stabilization of a discernible symptom) or physiologically (e.g., stabilization of a physical parameter) or both. In yet another embodiment, "treating" or "treatment" includes delaying or preventing the onset of the disease or disorder.

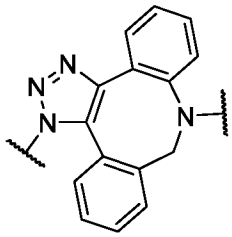
[0091] As used herein, the term "therapeutically effective amount" or "effective amount" refers to an amount of an antibody or composition that when administered to a subject is effective to treat a disease or disorder. In some embodiments, a therapeutically effective

amount or effective amount refers to an amount of an antibody or composition that when administered to a subject is effective to prevent or ameliorate a disease or the progression of the disease, or result in amelioration of symptoms.

[0092] As used herein, **the term "inhibits growth" (e.g. referring to cells, such as tumor** cells) is intended to include any measurable decrease in cell growth (*e.g.*, tumor cell growth) when contacted with a folate receptor alpha (FOLR1) antibody, as compared to the growth of the same cells not in contact with a FOLR1 antibody. In some embodiments, growth may be inhibited by at least about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 99%, or 100%. The decrease in cell growth can occur by a variety of mechanisms, including but not limited to antibody internalization, apoptosis, necrosis, and/or effector function-mediated activity.

[0093] **As used herein, the term "subject" means a mammalian subject. Exemplary subjects** include, but are not limited to humans, monkeys, dogs, cats, mice, rats, cows, horses, camels, avians, goats, and sheep. In certain embodiments, the subject is a human. In some embodiments, the subject has a disease that can be treated or diagnosed with an antibody provided herein. In some embodiments, the disease is gastric carcinoma, colorectal carcinoma, renal cell carcinoma, cervical carcinoma, non-small cell lung carcinoma, ovarian cancer, breast cancer, triple-negative breast cancer, endometrial cancer, prostate cancer, and/or a cancer of epithelial origin.

[0094] In some chemical structures illustrated herein, certain substituents, chemical groups, and atoms are depicted with a curvy/wavy line (e.g., ) that intersects a bond or bonds to indicate the atom through which the substituents, chemical groups, and atoms are bonded. For example, in some structures, such as but not limited to , , or , this curvy/wavy line indicates the atoms in the backbone of a conjugate or linker-payload structure to which the illustrated chemical entity is

bonded. In some structures, such as but not limited to , this curvy/wavy line indicates the atoms in the antibody or antibody fragment as well as the atoms in the backbone of a conjugate or linker-payload structure to which the illustrated chemical entity is bonded.

[0095] The term "site-specific" refers to a modification of a polypeptide at a predetermined sequence location in the polypeptide. The modification is at a single, predictable residue of the polypeptide with little or no variation. In particular embodiments, a modified amino acid is introduced at that sequence location, for instance recombinantly or synthetically. Similarly, a moiety can be "site-specifically" linked to a residue at a particular sequence location in the polypeptide. In certain embodiments, a polypeptide can comprise more than one site-specific modification.

2. Conjugates

[0096] Provided herein are conjugates of antibodies to folate receptor alpha (FOLR1 or FolRα). The conjugates comprise an antibody to FOLR1 covalently linked directly or indirectly, via a linker, to a payload. In certain embodiments, the antibody is linked to one payload. In further embodiments, the antibody is linked to more than one payload. In certain embodiments, the antibody is linked to two, three, four, five, six, seven, eight, or more payloads.

[0097] The payload can be any payload deemed useful by the practitioner of skill. In certain embodiments, the payload is a therapeutic moiety. In certain embodiments, the payload is a diagnostic moiety, *e.g.* a label. Useful payloads are described in the sections and examples below.

[0098] The linker can be any linker capable of forming at least one bond to the antibody and at least one bond to a payload. Useful linkers are described the sections and examples below.

[0099] In the conjugates provided herein, the antibody can be any antibody with binding specificity for folate receptor alpha (FOLR1 or FolRα). The FOLR1 can be from any species. In certain embodiments, the FOLR1 is a vertebrate FOLR1. In certain embodiments, the FOLR1 is a mammalian FOLR1. In certain embodiments, the FOLR1 is human FOLR1. In

certain embodiments, the FOLR1 is mouse FOLR1. In certain embodiments, the FOLR1 is cynomolgus FOLR1.

[00100] In certain embodiments, the antibody to folate receptor alpha (FOLR1 or FolRα) competes with an antibody described herein for binding. In certain embodiments, the antibody to FOLR1 binds to the same epitope as an antibody described herein.

[00101] The antibody is typically a protein comprising multiple polypeptide chains. In certain embodiments, the antibody is a heterotetramer comprising two identical light (L) chains and two identical heavy (H) chains. Each light chain can be linked to a heavy chain by one covalent disulfide bond. Each heavy chain can be linked to the other heavy chain by one or more covalent disulfide bonds. Each heavy chain and each light chain can also have one or more intrachain disulfide bonds. As is known to those of skill in the art, each heavy chain typically comprises a variable domain (V_H) followed by a number of constant domains. Each light chain typically comprises a variable domain at one end (V_L) and a constant domain. As is known to those of skill in the art, antibodies typically have selective affinity for their target molecules, *i.e.* antigens.

[00102] The antibodies provided herein can have any antibody form known to those of skill in the art. They can be full-length, or fragments. Exemplary full length antibodies include IgA, IgA1, IgA2, IgD, IgE, IgG, IgG1, IgG2, IgG3, IgG4, IgM, etc. Exemplary fragments include Fv, Fab, Fc, scFv, scFv-Fc, etc.

[00103] In certain embodiments, the antibody of the conjugate comprises one, two, three, four, five, or six of the CDR sequences described herein. In certain embodiments, the antibody of the conjugate comprises a heavy chain variable domain (V_H) described herein. In certain embodiments, the antibody of the conjugate comprises a light chain variable domain (V_L) described herein. In certain embodiments, the antibody of the conjugate comprises a heavy chain variable domain (V_H) described herein and a light chain variable domain (V_L) described herein. In certain embodiments, the antibody of the conjugate comprises a paired heavy chain variable domain and a light chain variable domain described herein (V_H - V_L pair).

[00104] In certain embodiments, the antibody of the conjugate comprises any of the amino acid sequences of the antibodies described above. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 10 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 9 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino

acid sequences above with up to 8 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 7 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 6 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 5 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 4 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 3 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 2 amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 1 conservative amino acid substitution. In some embodiments, the amino acid substitutions are conservative amino acid substitutions. For example, in certain embodiments, the antibody comprises any of the amino acid sequences above with up to 10 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 9 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 8 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 7 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 6 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 5 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 4 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 3 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 2 conservative amino acid substitutions. In certain embodiments, the antibody comprises any of the amino acid sequences above with up to 1 conservative amino acid substitution.

[00105] In certain embodiments, the antibody conjugate can be formed from an antibody that comprises one or more reactive groups. In certain embodiments, the antibody conjugate can be formed from an antibody comprising all naturally encoded amino acids. Those of skill in the art will recognize that several naturally encoded amino acids include reactive groups capable of conjugation to a payload or to a linker. These reactive groups include cysteine side chains, lysine side chains, and amino-terminal groups. In these embodiments, the antibody

conjugate can comprise a payload or linker linked to the residue of an antibody reactive group. In these embodiments, the payload precursor or linker precursor comprises a reactive group capable of forming a bond with an antibody reactive group. Typical reactive groups include maleimide groups, activated carbonates (including but not limited to, p-nitrophenyl ester), activated esters (including but not limited to, N-hydroxysuccinimide, p-nitrophenyl ester, and aldehydes). Particularly useful reactive groups include maleimide and succinimide, for instance N-hydroxysuccinimide, for forming bonds to cysteine and lysine side chains. Further reactive groups are described in the sections and examples below.

[00106] In further embodiments, the antibody comprises one or more modified amino acids having a reactive group, as described herein. Typically, the modified amino acid is not a naturally encoded amino acid. These modified amino acids can comprise a reactive group useful for forming a covalent bond to a linker precursor or to a payload precursor. One of skill in the art can use the reactive group to link the polypeptide to any molecular entity capable of forming a covalent bond to the modified amino acid. Thus, provided herein are conjugates comprising an antibody comprising a modified amino acid residue linked to a payload directly or indirectly via a linker. Exemplary modified amino acids are described in the sections below. Generally, the modified amino acids have reactive groups capable of forming bonds to linkers or payloads with complementary reactive groups.

[00107] The non-natural amino acids are positioned at select locations in a polypeptide chain of the antibody. These locations were identified as providing optimum sites for substitution with the non-natural amino acids. Each site is capable of bearing a non-natural amino acid with optimum structure, function and/or methods for producing the antibody.

[00108] In certain embodiments, a site-specific position for substitution provides an antibody that is stable. Stability can be measured by any technique apparent to those of skill in the art.

[00109] In certain embodiments, a site-specific position for substitution provides an antibody that has optimal functional properties. For instance, the antibody can show little or no loss of binding affinity for its target antigen compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced binding compared to an antibody without the site-specific non-natural amino acid.

[00110] In certain embodiments, a site-specific position for substitution provides an antibody that can be made advantageously. For instance, in certain embodiments, the antibody

shows advantageous properties in its methods of synthesis, discussed below. In certain embodiments, the antibody can show little or no loss in yield in production compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced yield in production compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show little or no loss of tRNA suppression compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced tRNA suppression in production compared to an antibody without the site-specific non-natural amino acid.

[00111] In certain embodiments, a site-specific position for substitution provides an antibody that has advantageous solubility. In certain embodiments, the antibody can show little or no loss in solubility compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced solubility compared to an antibody without the site-specific non-natural amino acid.

[00112] In certain embodiments, a site-specific position for substitution provides an antibody that has advantageous expression. In certain embodiments, the antibody can show little or no loss in expression compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced expression compared to an antibody without the site-specific non-natural amino acid.

[00113] In certain embodiments, a site-specific position for substitution provides an antibody that has advantageous folding. In certain embodiments, the antibody can show little or no loss in proper folding compared to an antibody without the site-specific non-natural amino acid. In certain embodiments, the antibody can show enhanced folding compared to an antibody without the site-specific non-natural amino acid.

[00114] In certain embodiments, a site-specific position for substitution provides an antibody that is capable of advantageous conjugation. As described below, several non-natural amino acids have side chains or functional groups that facilitate conjugation of the antibody to a second agent, either directly or via a linker. In certain embodiments, the antibody can show enhanced conjugation efficiency compared to an antibody without the same or other non-natural amino acids at other positions. In certain embodiments, the antibody can show enhanced conjugation yield compared to an antibody without the same or other non-natural amino acids at other positions. In certain embodiments, the antibody can show enhanced conjugation

specificity compared to an antibody without the same or other non-natural amino acids at other positions.

[00115] The one or more non-natural amino acids are located at selected site-specific positions in at least one polypeptide chain of the antibody. The polypeptide chain can be any polypeptide chain of the antibody without limitation, including either light chain or either heavy chain. The site-specific position can be in any domain of the antibody, including any variable domain and any constant domain.

[00116] In certain embodiments, the antibodies provided herein comprise one non-natural amino acid at a site-specific position. In certain embodiments, the antibodies provided herein comprise two non-natural amino acids at site-specific positions. In certain embodiments, the antibodies provided herein comprise three non-natural amino acids at site-specific positions. In certain embodiments, the antibodies provided herein comprise more than three non-natural amino acids at site-specific positions.

[00117] In certain embodiments, the antibodies provided herein comprise one or more non-natural amino acids each at a position selected from the group consisting of heavy chain or light chain residues HC-F404, HC-K121, HC-Y180, HC-F241, HC-221, LC-T22, LC-S7, LC-N152, LC-K42, LC-E161, LC-D170, HC-S136, HC-S25, HC-A40, HC-S119, HC-S190, HC-K222, HC-R19, HC-Y52, or HC-S70 according to the Kabat or Chothia or EU numbering scheme, or a post-translationally modified variant thereof. In these designations, HC indicates a heavy chain residue, and LC indicates a light chain residue.

[00118] In certain embodiments, provided herein are conjugates according to Formula (C1) or (C2):



(C1)



(C2)

or a pharmaceutically acceptable salt, solvate, stereoisomer, regioisomer, or tautomer thereof, wherein:

COMP is a residue of an anti-FOLR1 antibody;

PAY is a payload moiety;

W^1, **W^2**, **W^3**, **W^4**, and **W^5** are each independently a single bond, absent, or a divalent attaching group;

EG is absent, or an eliminator group;

each **RT** is a release trigger group, in the backbone of Formula (C1) or (C2) or bonded to **EG**, wherein each **RT** is optional;

HP is a single bond, absent, or a divalent hydrophilic group;

SG is a single bond, absent, or a divalent spacer group; and

R is hydrogen, a terminal conjugating group, or a divalent residue of a terminal conjugating group.

[00119] In some embodiments, a conjugate according to Formula (C1) or (C2) comprises **n** number of **PAY** moieties, wherein **n** is an integer from 1 to 8. In some embodiments, **n** is 2. In some embodiments, **n** is 3. In some embodiments, **n** is 4. In some embodiments, **n** is 5. In some embodiments, **n** is 6. In some embodiments, **n** is 7. In some embodiments, **n** is 8.

Attaching Groups

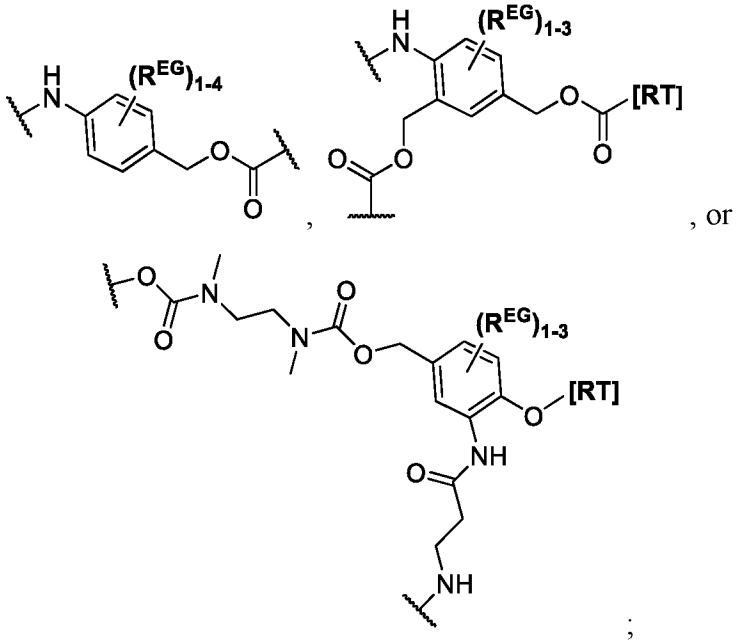
[00120] Attaching groups facilitate incorporation of eliminator groups, release trigger groups, hydrophobic groups, spacer groups, and/or conjugating groups into a compound. Useful attaching groups are known to, and are apparent to, those of skill in the art. Examples of useful attaching groups are provided herein. In certain embodiments, attaching groups are designated **W^1**, **W^2**, **W^3**, **W^4**, or **W^5**. In certain embodiments, an attaching group can comprise a divalent ketone, divalent ester, divalent ether, divalent amide, divalent amine, alkylene, arylene, sulfide, disulfide, carbonylene, or a combination thereof. In certain embodiments an attaching group can comprise –C(O)–, –O–, –C(O)NH–, –C(O)NH-alkyl–, –OC(O)NH–, –SC(O)NH–, –NH–, –NH-alkyl–, $–N(CH_3)CH_2CH_2N(CH_3)–$, –S–, –S-S–, $–OCH_2CH_2O–$, or the reverse (e.g. –NHC(O)–) thereof, or a combination thereof.

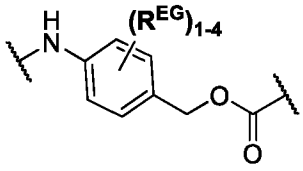
Eliminator Groups

[00121] Eliminator groups facilitate separation of a biologically active portion of a compound or conjugate described herein from the remainder of the compound or conjugate *in vivo* and/or *in vitro*. Eliminator groups can also facilitate separation of a biologically active portion of a compound or conjugate described herein in conjunction with a release trigger group. For example, the eliminator group and the release trigger group can react in a Releasing

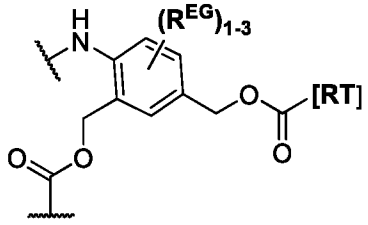
Reaction to release a biologically active portion of a compound or conjugate described herein from the compound or conjugate *in vivo* and/or *in vitro*. Upon initiation of the Releasing Reaction by the release trigger, the eliminator group cleaves the biologically active moiety, or a prodrug form of the biologically active moiety, and forms a stable, non-toxic entity that has no further effect on the activity of the biologically active moiety.

[00122] In certain embodiments, the eliminator group is designated **EG** herein. Useful eliminator groups include those described herein. In certain embodiments, the eliminator group is:

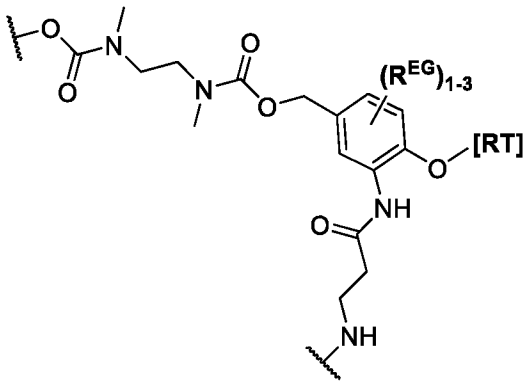


wherein $\mathbf{R}^{EG}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, $-NO_2$, -CN, fluoro, bromo, chloro, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In each structure, the phenyl ring can be bound to one, two, three, or in some cases, four R^{EG} groups. In the second and third structures, those of skill will recognize that EG is bonded to an RT that is not within the backbone of formula (C1) as indicated in the above description of formula (C1). In some embodiments, R^{EG} is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In further embodiments, R^{EG} is selected from the group consisting of hydrogen, $-NO_2$, -CN, fluoro, bromo, and chloro. In certain embodiments, the eliminator group is 

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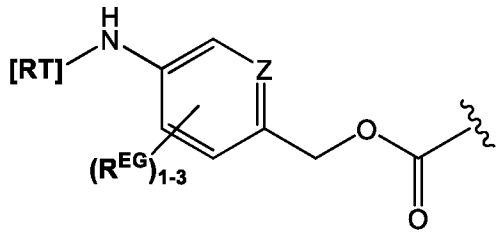


embodiments, the eliminator group is . In certain embodiments,

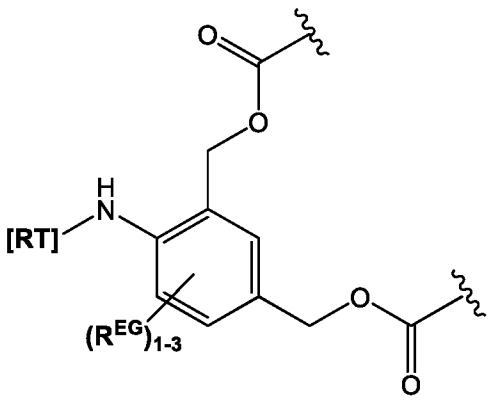


the eliminator group is .

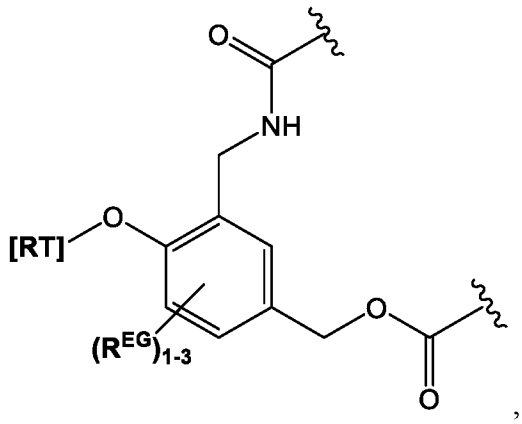
[00123] In some embodiments, the eliminator group is:



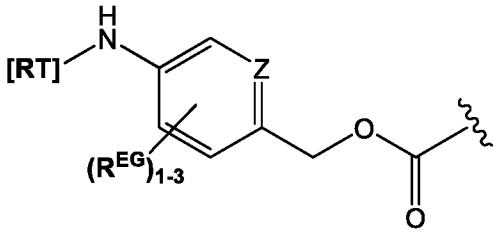
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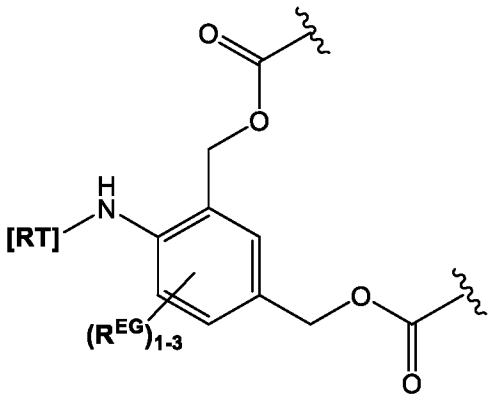
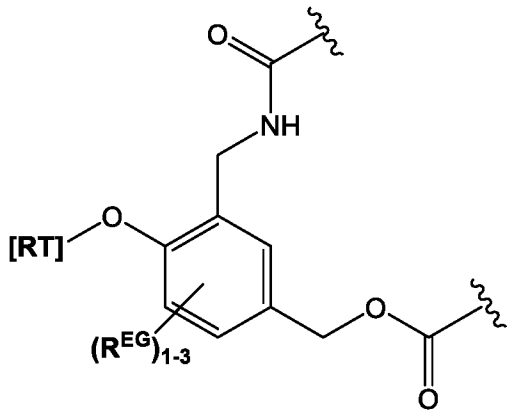
, or



,

wherein Z may be CH or N, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, $-NO_2$, -CN, fluoro, bromo, chloro, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In each structure, the phenyl ring can be bound to one, two, three, or in some cases, four R^{EG} groups. In the first and second structures, those of skill will recognize that EG is bonded to an RT that is not within the backbone of formula (C1) as indicated in the above description of formula (C1). In some embodiments, R^{EG} is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In further embodiments, R^{EG} is selected from the group consisting of hydrogen, $-NO_2$, -CN, fluoro, bromo, and chloro. In some embodiments, each R^{EG} in the EG is hydrogen. In certain embodiments, the eliminator group is 

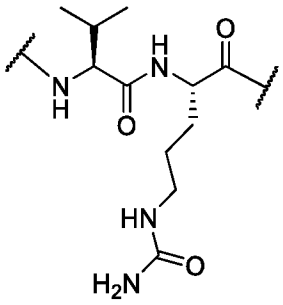
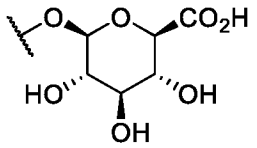
. In certain embodiments, the

eliminator group is  . In certain embodiments, the eliminator group is  .

Release Trigger Groups

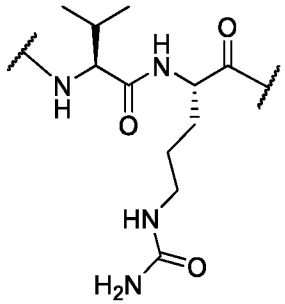
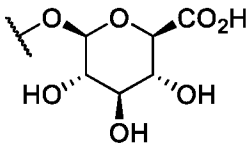
[00124] Release trigger groups facilitate separation of a biologically active portion of a compound or conjugate described herein from the remainder of the compound or conjugate *in vivo* and/or *in vitro*. Release trigger groups can also facilitate separation of a biologically active portion of a compound or conjugate described herein in conjunction with an eliminator group. For example, the eliminator group and the release trigger group can react in a Releasing Reaction to release a biologically active portion of a compound or conjugate described herein from the compound or conjugate *in vivo* and/or *in vitro*. In certain embodiment, the release trigger can act through a biologically-driven reaction with high tumor:nontumor specificity, such as the proteolytic action of an enzyme overexpressed in a tumor environment.

[00125] In certain embodiments, the release trigger group is designated **RT** herein. In certain embodiments, **RT** is divalent and bonded within the backbone of formula (C1). In other embodiments, **RT** is monovalent and bonded to **EG** as depicted above. Useful release trigger groups include those described herein. In certain embodiments, the release trigger group comprises a residue of a natural or non-natural amino acid or residue of a sugar ring. In certain embodiments, the release trigger group is:

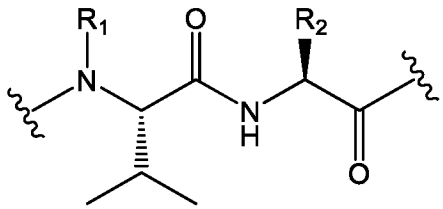



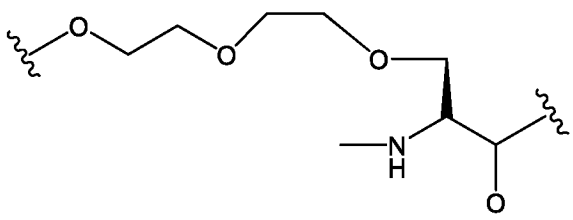
or .

Those of skill will recognize that the first structure is divalent and can be bonded within the backbone of Formula (C1) or as depicted in Formula (C2), and that the second structure is monovalent and can be bonded to **EG** as depicted in formula (C1) above.

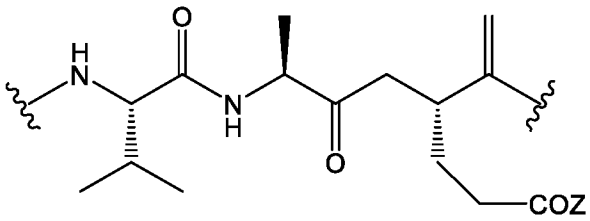
In certain embodiments, the release trigger group is . In certain embodiments, the release trigger group is .

[00126] In some embodiments, the release trigger group is a protease-cleavable R_1-Val-X peptide having the structure of:

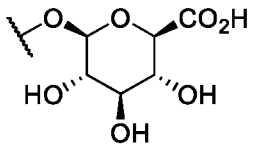


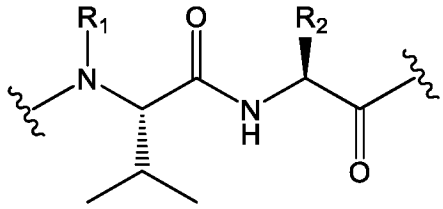
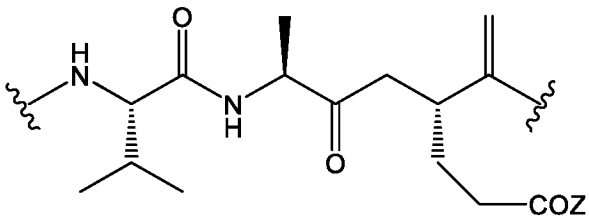
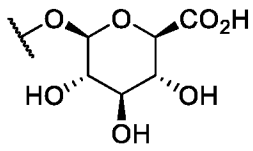


wherein R_1 is H or and R_2 is CH_3, $CH_2CH_2CO_2H$, or $(CH_2)_3NHCONH_2$; a legumain-cleavable Ala-Ala-Asn or Ala-Ala-Asp peptide having the structure of:



where Z is OH or NH_2; or a β-glucuronidase-cleavable β-glucuronide having the structure of:



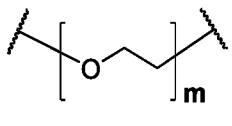
Those of skill will recognize that  and  are divalent structures and can be bonded within the backbone of Formula (C1) or as depicted in Formula (C2). The structure  is monovalent and can be bonded to **EG** as depicted in formula (C1) above.

Hydrophilic Groups

[00127] Hydrophilic groups facilitate increasing the hydrophilicity of the compounds described herein. It is believed that increased hydrophilicity allows for greater solubility in

aqueous solutions, such as aqueous solutions found in biological systems. Hydrophilic groups can also function as spacer groups, which are described in further detail herein.

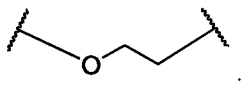
[00128] In certain embodiments, the hydrophilic group is designated **HP** herein. Useful hydrophilic groups include those described herein. In certain embodiments, the hydrophilic group is a divalent poly(ethylene glycol). In certain embodiments, the hydrophilic group is a divalent poly(ethylene glycol) according to the formula:



;

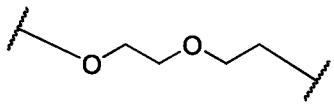
wherein **m** is an integer from 1 to 13, optionally 1 to 4, optionally 2 to 4, or optionally 4 to 8.

[00129] In some embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



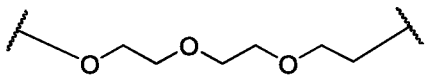
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[00130] In some other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



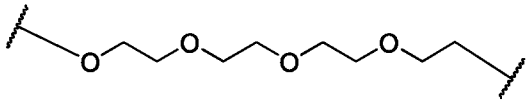
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[00131] In other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



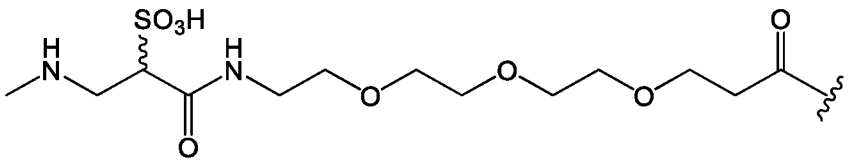
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[00132] In other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



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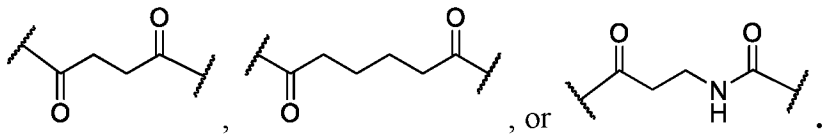
[00133] In some embodiments, the hydrophilic group can bear a chain-presented sulfonic acid having the formula:



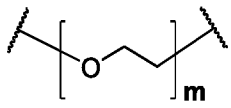
Spacer Groups

[00134] Spacer groups facilitate spacing of the conjugating group from the other groups of the compounds described herein. This spacing can lead to more efficient conjugation of the compounds described herein to a second compound as well as more efficient cleavage of the active catabolite. The spacer group can also stabilize the conjugating group and lead to improved overall antibody-drug conjugate properties.

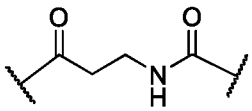
[00135] In certain embodiments, the spacer group is designated **SP** herein. Useful spacer groups include those described herein. In certain embodiments, the spacer group is:



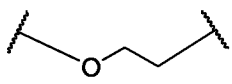
In certain embodiments, the spacer group, $\mathbf{W^4}$, and the hydrophilic group combine to form a divalent poly(ethylene glycol) according to the formula:



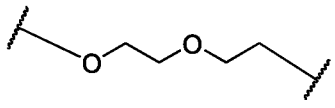
wherein **m** is an integer from 1 to 13, optionally 1 to 4, optionally 2 to 4, or optionally 4 to 8.

[00136] In some embodiments, the **SP** is .

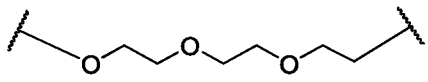
[00137] In some embodiments, the divalent poly(ethylene glycol) has the following formula:



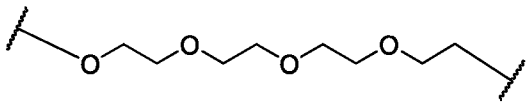
[00138] In some other embodiments, the divalent poly(ethylene glycol) has the following formula:



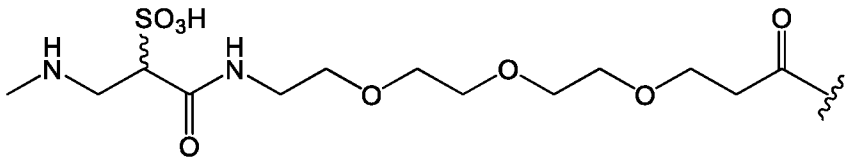
[00139] In other embodiments, the divalent poly(ethylene glycol) has the following formula:



[00140] In other embodiments, the divalent poly(ethylene glycol) has the following formula:

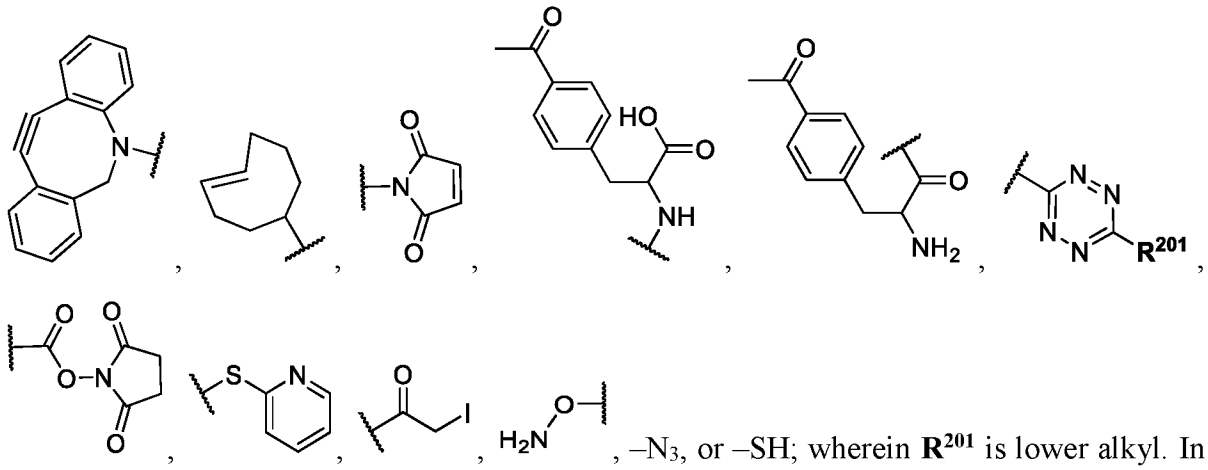


[00141] In some embodiments, the hydrophilic group can bear a chain-presented sulfonic acid having the formula:



Conjugating Groups and Residues Thereof

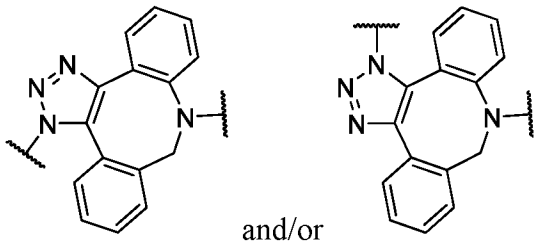
[00142] Conjugating groups facilitate conjugation of the payloads described herein to a second compound, such as an antibody described herein. In certain embodiments, the conjugating group is designated **R** herein. Conjugating groups can react via any suitable reaction mechanism known to those of skill in the art. In certain embodiments, a conjugating group reacts through a [3+2] alkyne-azide cycloaddition reaction, inverse-electron demand Diels-Alder ligation reaction, thiol-electrophile reaction, or carbonyl-oxyamine reaction, as described in detail herein. In certain embodiments, the conjugating group comprises an alkyne, strained alkyne, tetrazine, thiol, para-acetyl-phenylalanine residue, oxyamine, maleimide, or azide. In certain embodiments, the conjugating group is:



, –N_3, or –SH; wherein $\mathbf{R^{201}}$ is lower alkyl. In an embodiment, $\mathbf{R^{201}}$ is methyl, ethyl, or propyl. In an embodiment, $\mathbf{R^{201}}$ is methyl. Additional conjugating groups are described in, for example, U.S. Patent Publication No. 2014/0356385, U.S. Patent Publication No. 2013/0189287, U.S. Patent Publication No. 2013/0251783, U.S. Patent No. 8,703,936, U.S. Patent No. 9,145,361, U.S. Patent No. 9,222,940, and U.S. Patent No. 8,431,558.

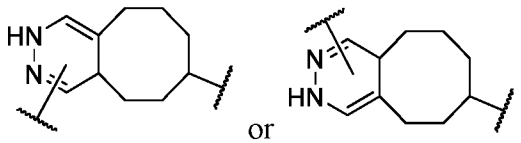
[00143] After conjugation, a divalent residue of the conjugating group is formed and is bonded to the residue of a second compound. The structure of the divalent residue is determined by the type of conjugation reaction employed to form the conjugate.

[00144] In certain embodiments when a conjugate is formed through a [3+2] alkyne-azide cycloaddition reaction, the divalent residue of the conjugating group comprises a triazole ring or fused cyclic group comprising a triazole ring. In certain embodiment when a conjugate is formed through a strain-promoted [3+2] alkyne-azide cycloaddition (SPAAC) reaction, the divalent residue of the conjugating group is:

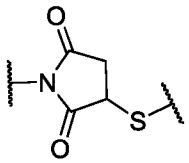


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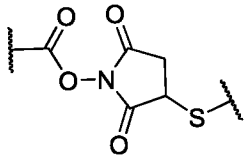
[00145] In certain embodiments when a conjugate is formed through a tetrazine inverse electron demand Diels-Alder ligation reaction, the divalent residue of the conjugating group comprises a fused bicyclic ring having at least two adjacent nitrogen atoms in the ring. In certain embodiments when a conjugate is formed through a tetrazine inverse electron demand Diels-Alder ligation reaction, the divalent residue of the conjugating group is:



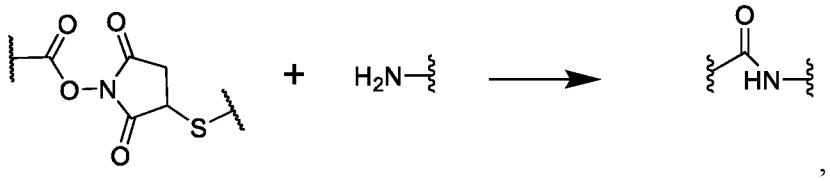
[00146] In certain embodiments when a conjugate is formed through a thiol-maleimide reaction, the divalent residue of the conjugating group comprises succinimidylene and a sulfur linkage. In certain embodiments when a conjugate is formed through a thiol-maleimide reaction, the divalent residue of the conjugating group is:



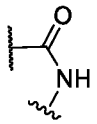
[00147] In certain embodiments, a conjugate is formed through a thiol-N-hydroxysuccinimide reaction using the following group:



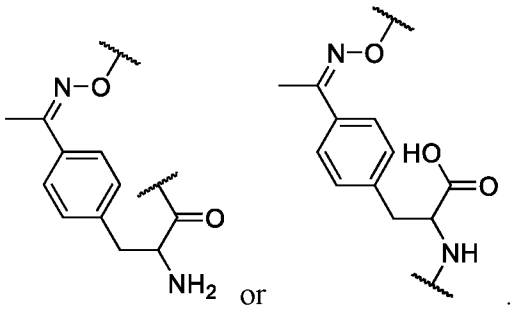
The reaction involved for formation of the conjugate comprises the following step:



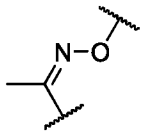
and the resulting divalent residue of the conjugating group is:



[00148] In certain embodiments when a conjugate is formed through a carbonyl-oxyamine reaction, the divalent residue of the conjugating group comprises a divalent residue of a non-natural amino acid. In certain embodiments when a conjugate is formed through a carbonyl-oxyamine reaction, the divalent residue of the conjugating group is:

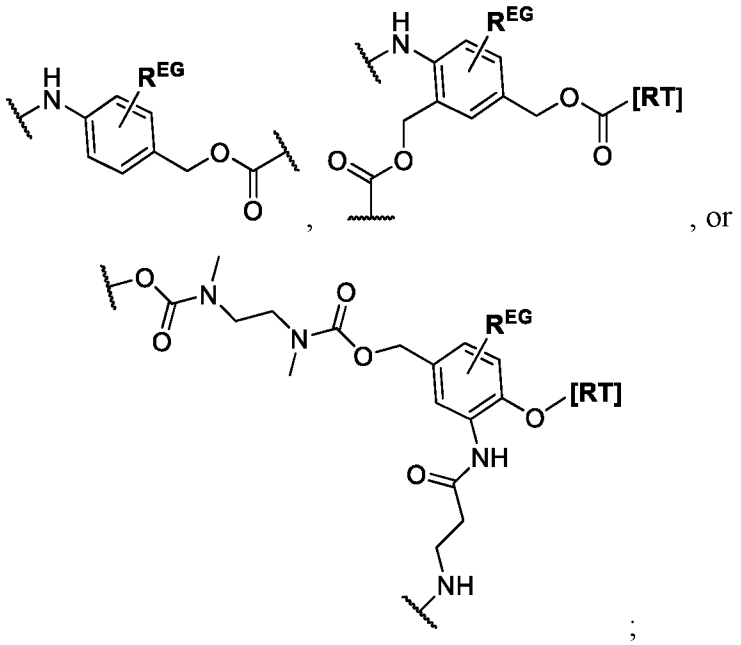


[00149] In certain embodiments when a conjugate is formed through a carbonyl-oxyamine reaction, the divalent residue of the conjugating group comprises an oxime linkage. In certain embodiments when a conjugate is formed through a carbonyl-oxyamine reaction, the divalent residue of the conjugating group is:



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[00150] In some embodiment, provided herein is a conjugate according to Formula (C1) or (C2) or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **EG** comprises phenylene, carboxylene, amine, or a combination thereof. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **EG** is:

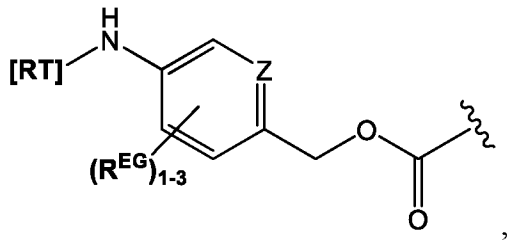


;

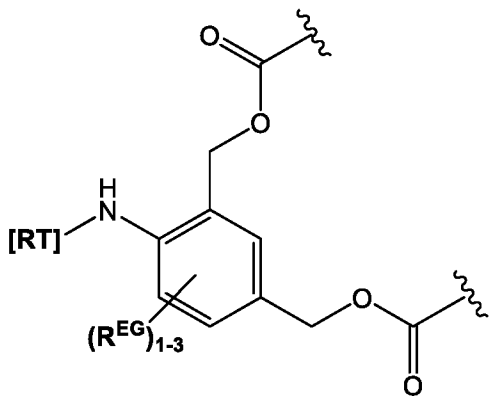
wherein $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, $-NO_2$, -CN, fluoro, bromo, chloro, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-,

alkylamino-C(O)- and dialkylaminoC(O)-. In each structure, the phenyl ring can be bound to one, two, three, or in some cases, four $\mathbf{R^{EG}}$ groups. In the second and third structures, those of skill will recognize that **EG** is bonded to an **RT** that is not within the backbone of Formula C1 as indicated in the above description of Formula C1. In some embodiments, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In further embodiments, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, $-NO_2$, -CN, fluoro, bromo, and chloro.

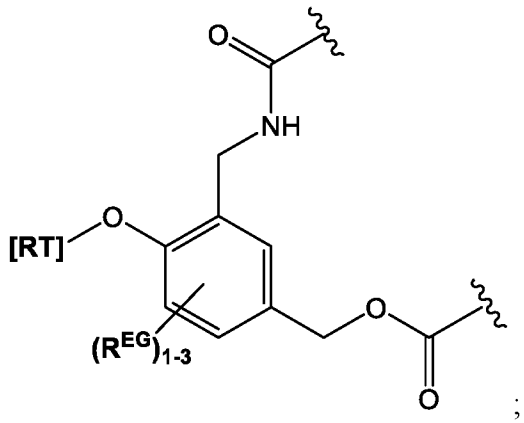
[00151] In some embodiments, provided herein is a conjugate according to Formula (C1) or (C2) or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **EG** comprises phenylene, carboxylene, amine, or a combination thereof. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **EG** is:



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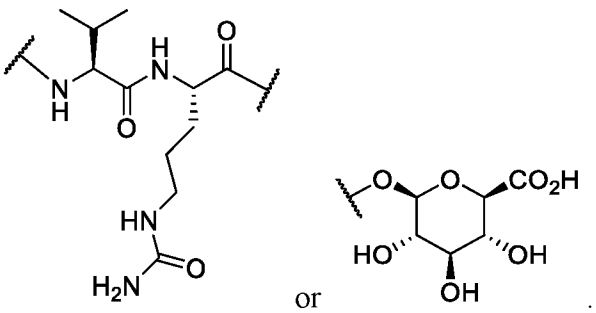


, or



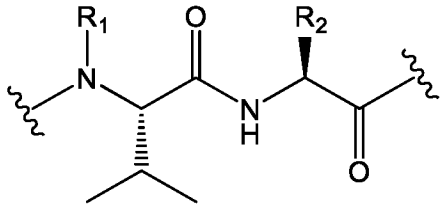
wherein Z may be CH or N, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, $-NO_2$, -CN, fluoro, bromo, chloro, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In each structure, the phenyl ring can be bound to one, two, three, or in some cases, four $\mathbf{R^{EG}}$ groups. In the second and third structures, those of skill will recognize that **EG** is bonded to an **RT** that is not within the backbone of Formula C1 as indicated in the above description of Formula C1. In some embodiments, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, alkyl, biphenyl, $-CF_3$, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In further embodiments, $\mathbf{R^{EG}}$ is selected from the group consisting of hydrogen, $-NO_2$, -CN, fluoro, bromo, and chloro. In some embodiments, each R^{EG} in the EG is hydrogen.

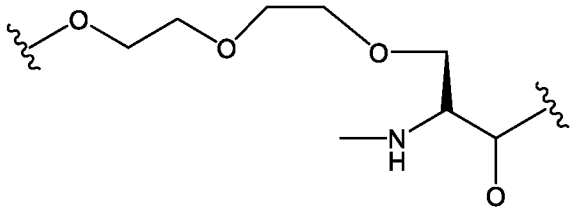
[00152] In some embodiments, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **RT** comprises a residue of a natural or non-natural amino acid or a residue of a sugar. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **RT** is:



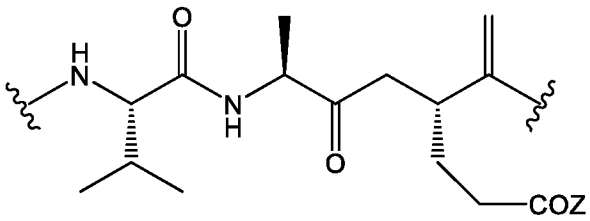
Those of skill will recognize that the first structure is divalent and can be bonded within the backbone as depicted in Formula (C2), and that the second structure is monovalent and can be bonded to **EG** as depicted in Formula (C1) above.

[00153] In some embodiments, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **RT** comprises a residue of a natural or non-natural amino acid or a residue of a sugar. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **RT** is:

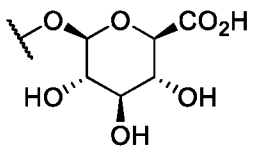


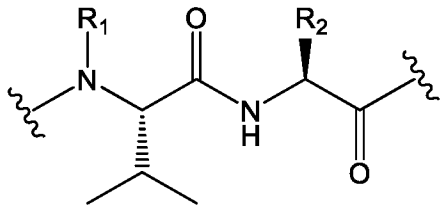
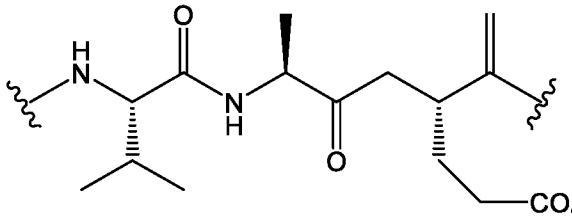
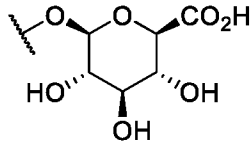


wherein R_1 is H or and R_2 is CH_3, $CH_2CH_2CO_2H$, or $(CH_2)_3NHCONH_2$; a legumain-cleavable Ala-Ala-Asn or Ala-Ala-Asp peptide having the structure of:

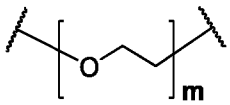


where Z is OH or NH_2; or a β-glucuronidase-cleavable β-glucuronide having the structure of:



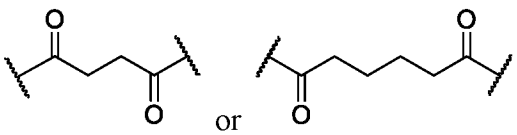
Those of skill will recognize that  and  are divalent structures and can be bonded within the backbone of Formula (C1) or as depicted in Formula (C2). The structure  is monovalent and can be bonded to **EG** as depicted in formula (C1) above.

[00154] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **HP** comprises poly(ethylene glycol). In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **HP** is:



wherein **m** is an integer from 1 to 13.

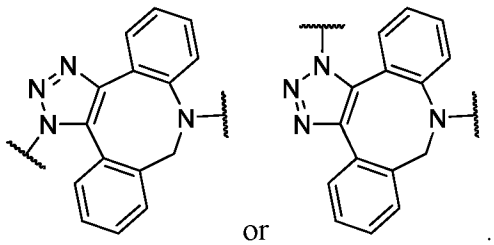
[00155] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **SG** comprises C_1-C_{10} alkylene, C_4-C_6 alkylene, carbonylene, or combination thereof. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **SG** is:

 or .

[00156] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein $\mathbf{W^1}$, $\mathbf{W^2}$, $\mathbf{W^3}$, $\mathbf{W^4}$, and $\mathbf{W^5}$ are each independently a single bond, absent, or comprise a divalent

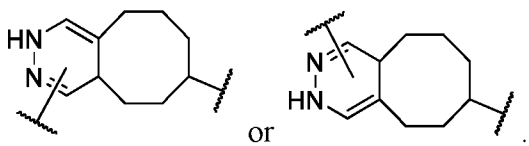
ketone, divalent ester, divalent ether, divalent amide, divalent amine, alkylene, arylene, sulfide, disulfide, carbonylene, or a combination thereof. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein $\mathbf{W^1}$, $\mathbf{W^2}$, $\mathbf{W^3}$, $\mathbf{W^4}$, and $\mathbf{W^5}$ are each independently a single bond, absent, or comprise –C(O)–, –O–, –C(O)NH–, —C(O)NH-alkyl–, –OC(O)NH–, –SC(O)NH–, –NH–, —NH–alkyl–, —$N(CH_3)CH_2CH_2N(CH_3)$–, –S–, –S-S–, $-OCH_2CH_2O-$, or a combination thereof.

[00157] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** comprises a triazole ring. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is a triazole ring or fused cyclic group comprising a triazole ring. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:



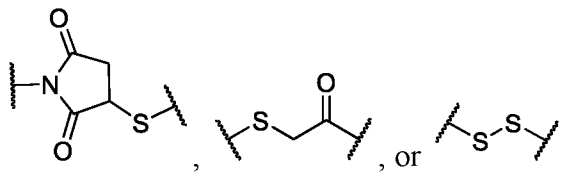
or .

[00158] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** comprises a fused bicyclic ring having at least two adjacent nitrogen atoms in the ring. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:

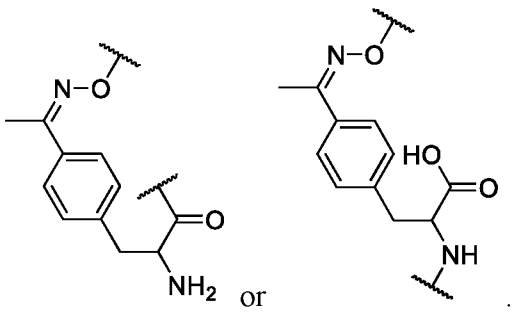


or .

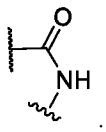
[00159] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** comprises a sulfur linkage. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:



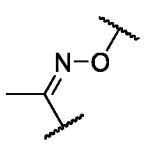
[00160] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** comprises a divalent residue of a non-natural amino acid. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:



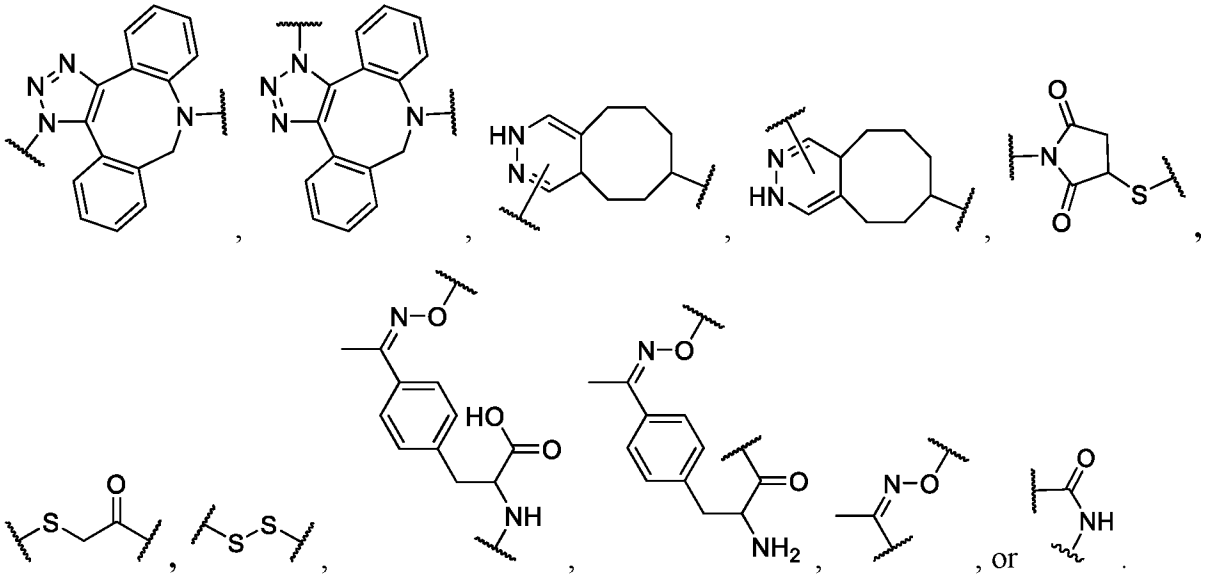
[00161] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein comprises an oxime linkage. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:



[00162] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein comprises an oxime linkage. In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:

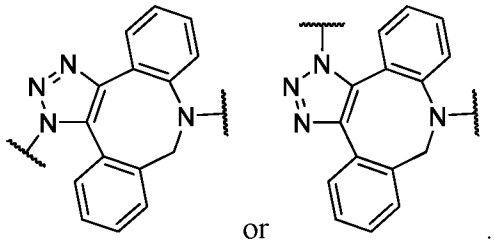


[00163] In an embodiment, provided herein is a conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **R** is:

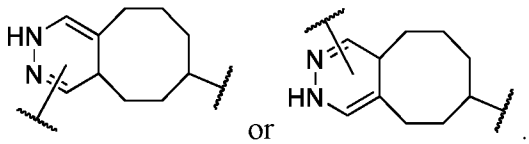


[00164] In an embodiment, provided herein is a compound according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **COMP** is a residue of any compound known to be useful for conjugation to a payload, described herein, and an optional linker, described herein. In an embodiment, provided herein is a compound according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof; wherein **COMP** is a residue of an antibody chain.

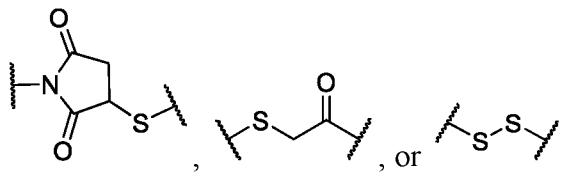
[00165] In an aspect, provided herein is an antibody conjugate comprising payload, described herein, and an optional linker, described herein, linked to an anti-FOLR1 antibody, wherein **COMP** is a residue of the antibody. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a triazole ring or fused cyclic group comprising a triazole ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:



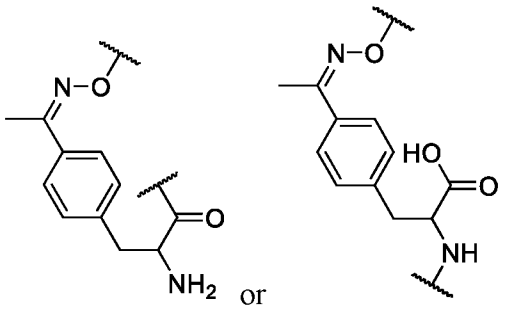
[00166] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a fused bicyclic ring, wherein the fused bicyclic ring has at least two adjacent nitrogen atoms in the ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:



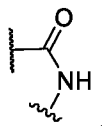
[00167] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** comprises a sulfur linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** is:



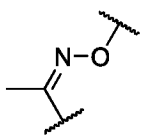
[00168] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** comprises a divalent residue of a non-natural amino acid. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** is:



[00169] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** is:

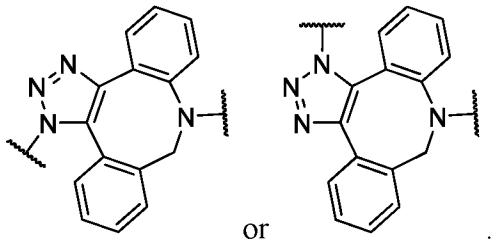


[00170] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the polypeptide; and **R** is:

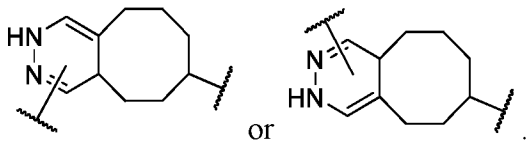


[00171] In an aspect, provided herein is an antibody conjugate comprising a payload, described herein, and an optional linker, described herein, linked to an antibody according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein **COMP** is a residue of the antibody. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a triazole ring or fused cyclic group comprising a triazole ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a

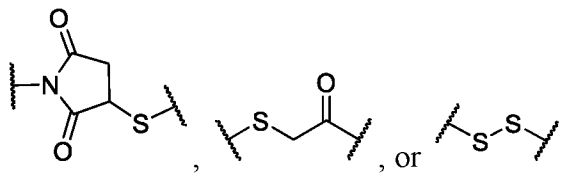
pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:



[00172] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a fused bicyclic ring, wherein the fused bicyclic ring has at least two adjacent nitrogen atoms in the ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:

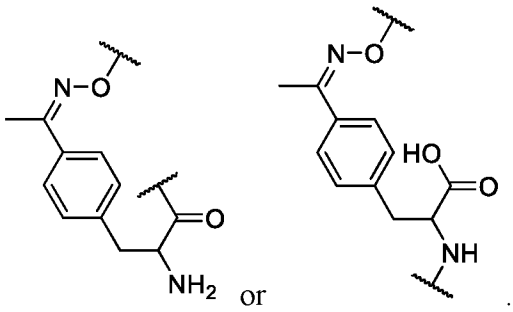


[00173] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a sulfur linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:

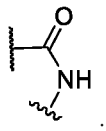


[00174] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises a divalent residue of a non-natural amino acid. In an embodiment, provided herein is an antibody conjugate according to

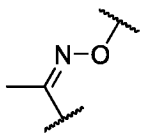
Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:



[00175] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:

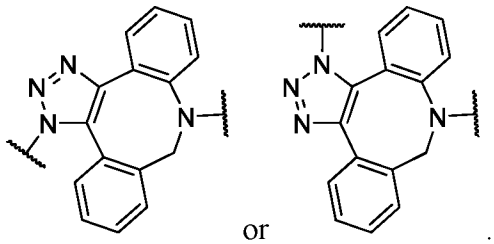


[00176] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody; and **R** is:

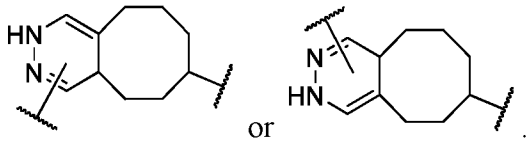


[00177] In an aspect, provided herein is an antibody conjugate comprising a payload, described herein, and an optional linker, described herein, linked to an antibody chain according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein **COMP** is a residue of the antibody chain. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP**

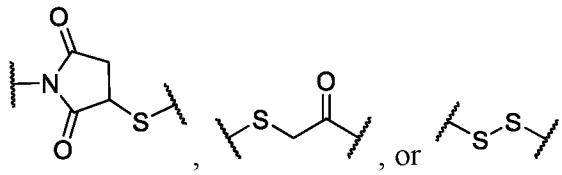
is a residue of the antibody chain; and **R** comprises a triazole ring or fused cyclic group comprising a triazole ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:



[00178] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** comprises a fused bicyclic ring, wherein the fused bicyclic ring has at least two adjacent nitrogen atoms in the ring. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:

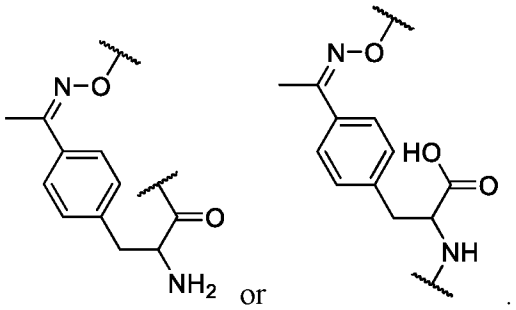


[00179] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** comprises a sulfur linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:

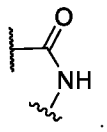


[00180] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** comprises a divalent residue

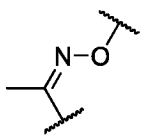
of a non-natural amino acid. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:



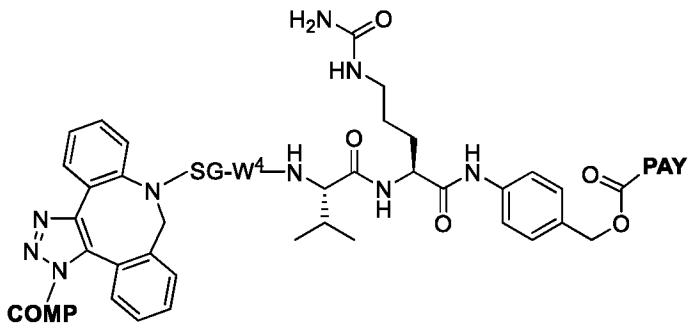
[00181] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:

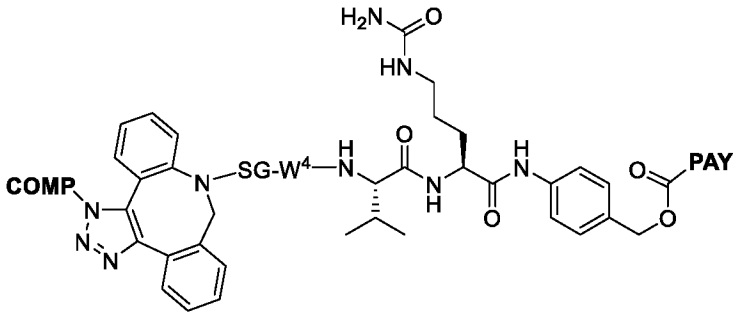


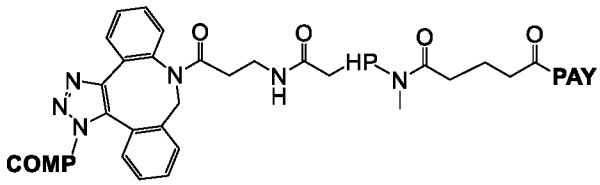
[00182] In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** comprises an oxime linkage. In an embodiment, provided herein is an antibody conjugate according to Formula (C1) or (C2), or a pharmaceutically acceptable salt, solvate, stereoisomer, or tautomer thereof, wherein: **COMP** is a residue of the antibody chain; and **R** is:

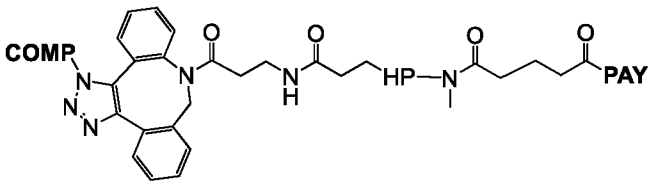


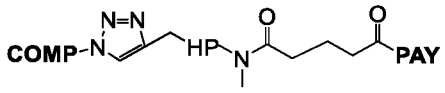
[00183] In an embodiment, provided herein is a conjugate according to any of the following formulas, where **COMP** indicates a residue of the anti-FOLR1 antibody and **PAY** indicates a payload moiety:

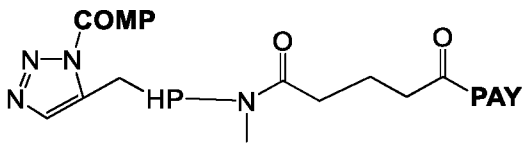

H2N
O
HN
SG-W4
H
N
O
N
H
O
N
H
O
PAY
O
O
N
N
N
COMP

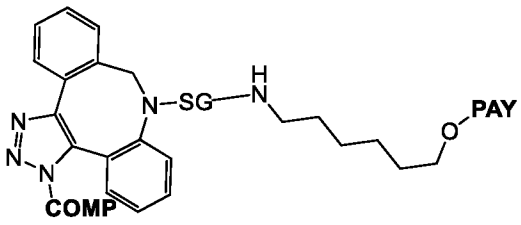

H2N
O
HN
COMP
N
N
N
SG-W4
H
N
O
N
H
O
N
H
O
PAY
O
O


O
O
O
O
N
N
N
N
H
HP
N
PAY
COMP

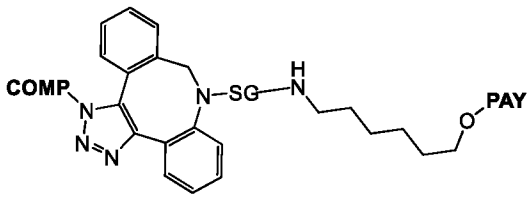

COMP
N
N
N
O
O
O
O
N
N
H
HP
N
PAY


N=N
COMP
N
HP
O
O
N
PAY


COMP
N
N
N
HP
O
O
N
PAY

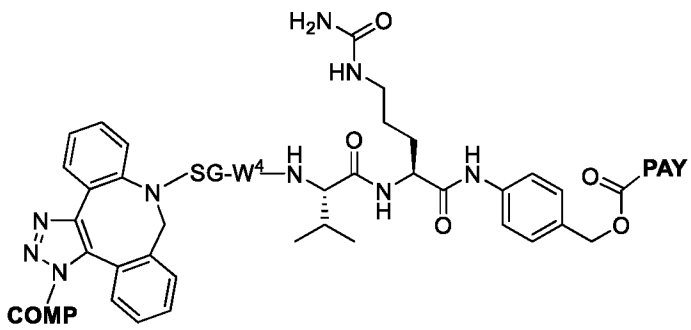


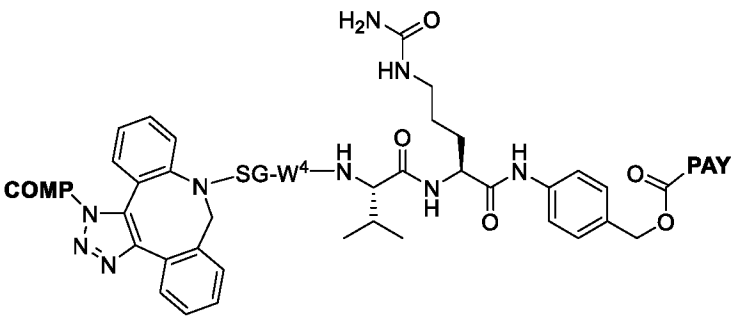
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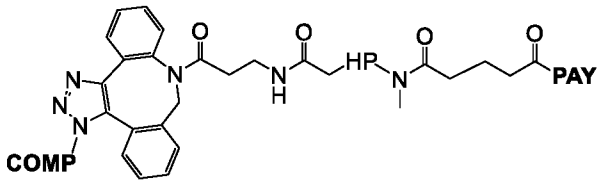


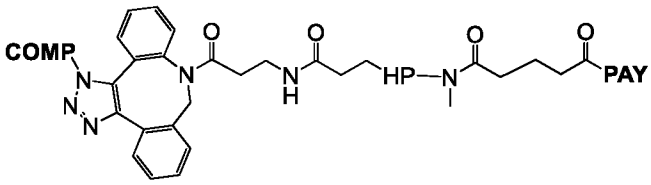
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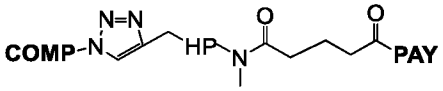
[00184] In an embodiment, provided herein is a conjugate according to any of the following formulas, where **COMP** indicates a residue of the anti-FOLR1 antibody and **PAY** indicates a payload moiety:

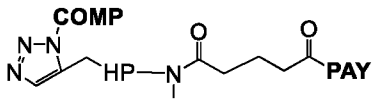


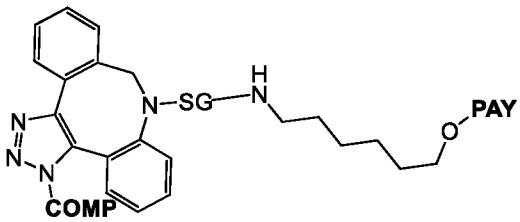




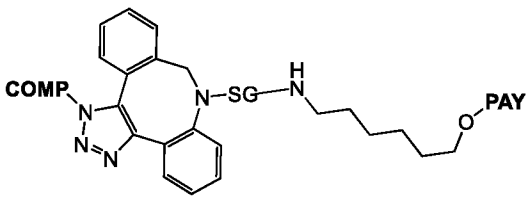






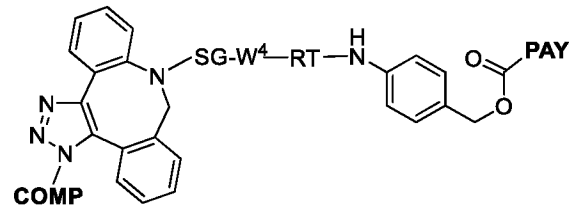


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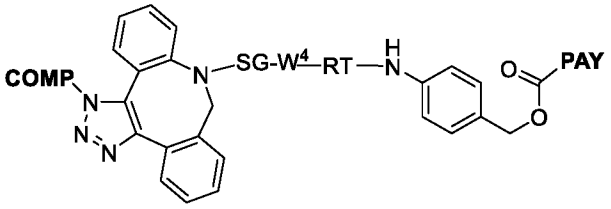


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[00185] In an embodiment, provided herein is a conjugate according to any of the following formulas, where **COMP** indicates a residue of the anti-FOLR1 antibody and **PAY** indicates a payload moiety:

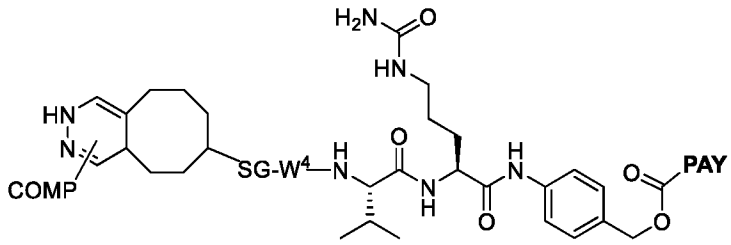


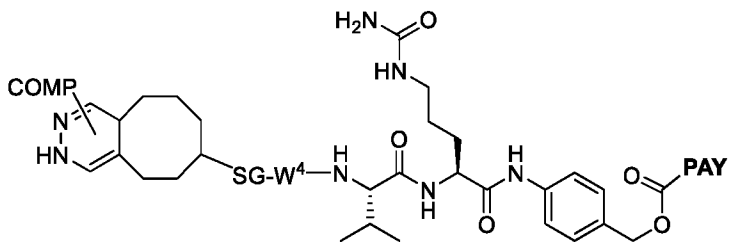
or

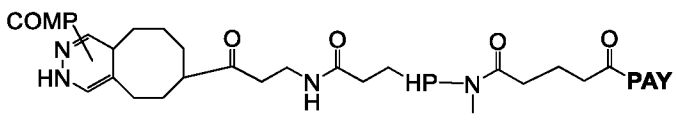


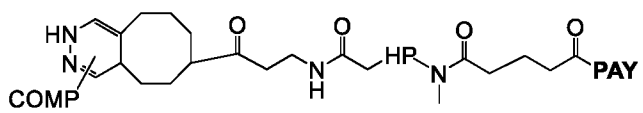
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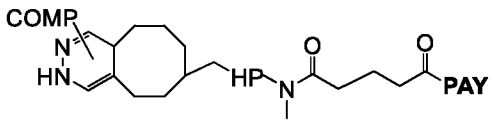
[00186] In an embodiment, provided herein is a conjugate according to any of the following formulas, where **COMP** indicates a residue of the anti-FOLR1 antibody and **PAY** indicates a payload moiety:

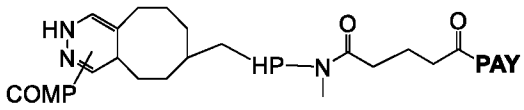


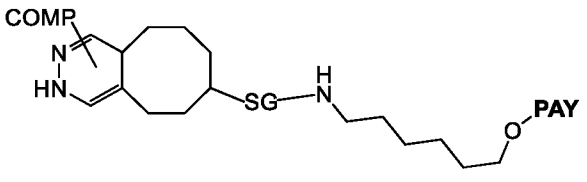




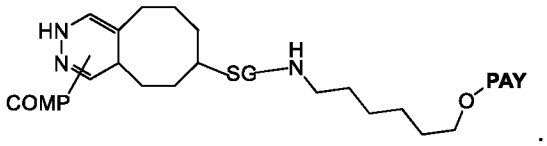






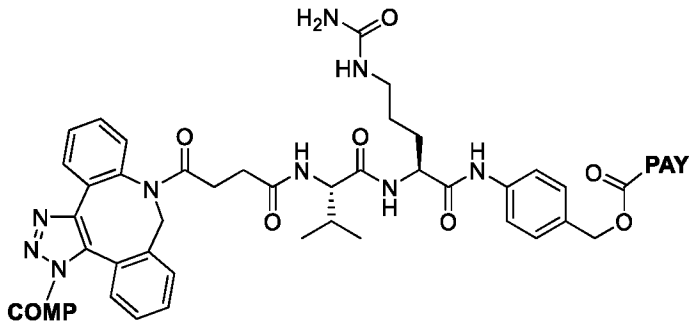


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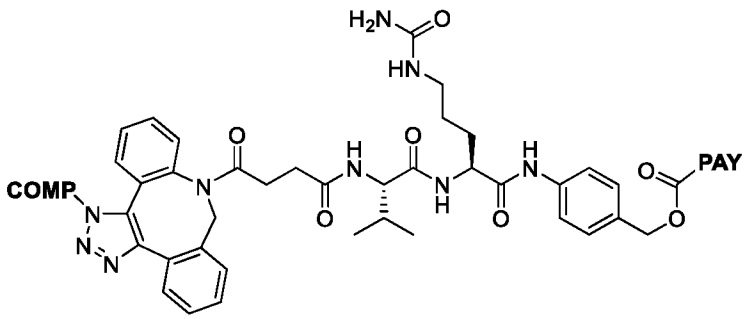


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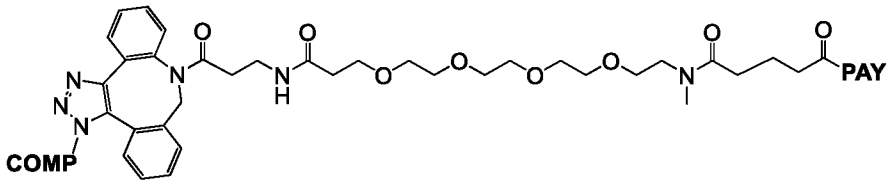
[00187] In an embodiment, provided herein is a conjugate according to any of Formulas 101a-104b, where **COMP** indicates a residue of the anti-FOLR1 antibody and **PAY** indicates a payload moiety:



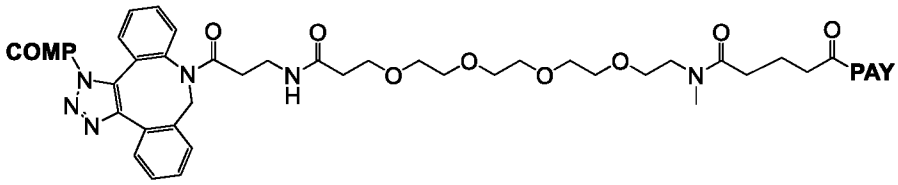
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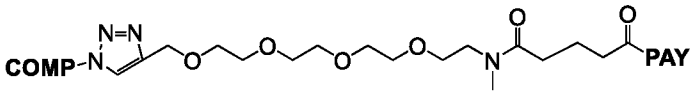
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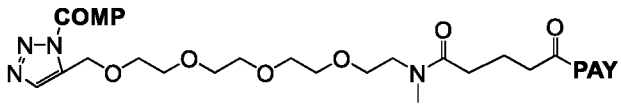
(102a)



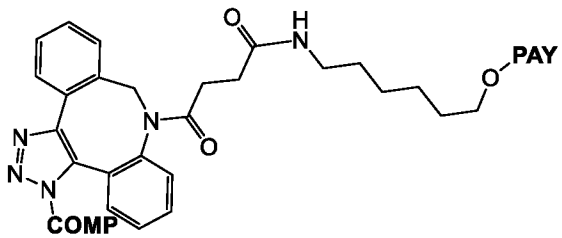
(102b)



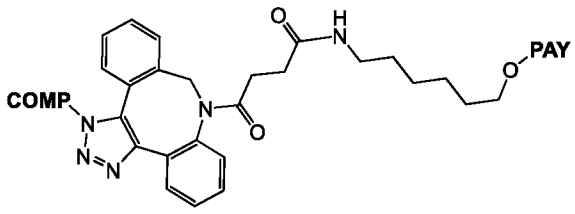
(103a)



(103b)



(104a)

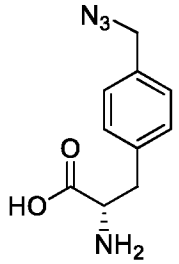


(104b).

[00188] In any of the foregoing embodiments, the conjugate comprises **n** number of **PAY** moieties, wherein **n** is an integer from 1 to 8. In some embodiments, **n** is 2. In some embodiments, **n** is 3. In some embodiments, **n** is 4. In some embodiments, **n** is 5. In some embodiments, **n** is 6. In some embodiments, **n** is 7. In some embodiments, **n** is 8.

[00189] In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at heavy chain position 404 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at heavy chain position 180 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at heavy chain position 241 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at heavy chain position 222 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at light chain position 7 according to the Kabat or Chothia numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (30), below, at light chain position 42 according to the Kabat or Chothia numbering system. In certain embodiments, **PAY** is selected from the group consisting of maytansine, hemiasterlin, amanitin, monomethyl auristatin F (MMAF), and monomethyl auristatin E (MMAE). In certain

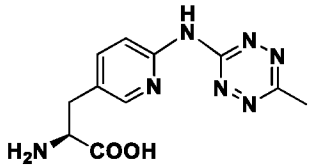
embodiments, the **PAY** is maytansine. In certain embodiments, **PAY** is hemiasterlin. In certain embodiments, **PAY** is amanitin. In certain embodiments, **PAY** is MMAF. In certain embodiments, **PAY** is MMAE.



(30)

[00190] In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at heavy chain position 404 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at heavy chain position 180 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at heavy chain position 241 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at heavy chain position 222 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at light chain position 7 according to the Kabat or Chothia numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a residue of the non-natural amino acid according to Formula (56), below, at light chain position 42 according to the Kabat or Chothia numbering system. In certain embodiments, **PAY** is selected from the group consisting of maytansine, hemiasterlin, amanitin, MMAF, and MMAE. In certain embodiments, the **PAY** is maytansine. In certain

embodiments, **PAY** is hemiasterlin. In certain embodiments, **PAY** is amanitin. In certain embodiments, **PAY** is MMAF. In certain embodiments, **PAY** is MMAE.



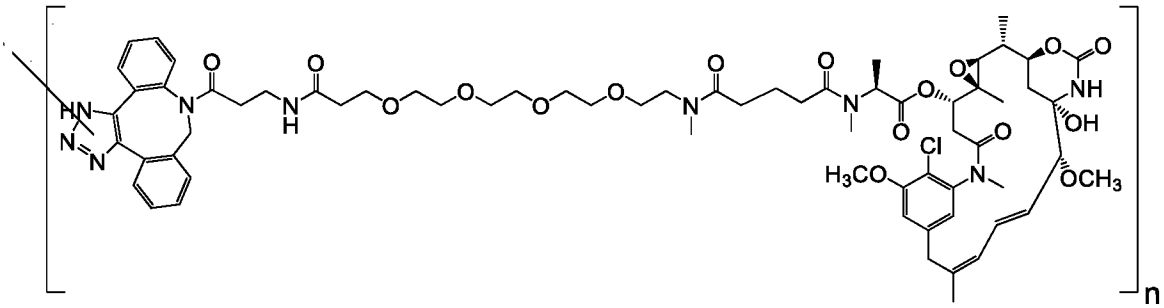
(56)

[00191] In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue of para-azido-L-phenylalanine. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates the non-natural amino acid residue para-azido-phenylalanine at heavy chain position 404 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue of para-azido-L-phenylalanine at heavy chain position 180 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue para-azido-L-phenylalanine at heavy chain position 241 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue para-azido-L-phenylalanine at heavy chain position 222 according to the EU numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue para-azido-L-phenylalanine at light chain position 7 according to the Kabat or Chothia numbering system. In particular embodiments, provided herein are anti-FOLR1 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue para-azido-L-phenylalanine at light chain position 42 according to the Kabat or Chothia numbering system. In certain embodiments, **PAY** is selected from the group consisting of maytansine, hemiasterlin, amanitin, MMAF, and MMAE. In certain embodiments, the **PAY** is maytansine. In certain embodiments, **PAY** is hemiasterlin. In certain embodiments, **PAY** is amanitin. In certain embodiments, **PAY** is MMAF. In certain embodiments, **PAY** is MMAE.

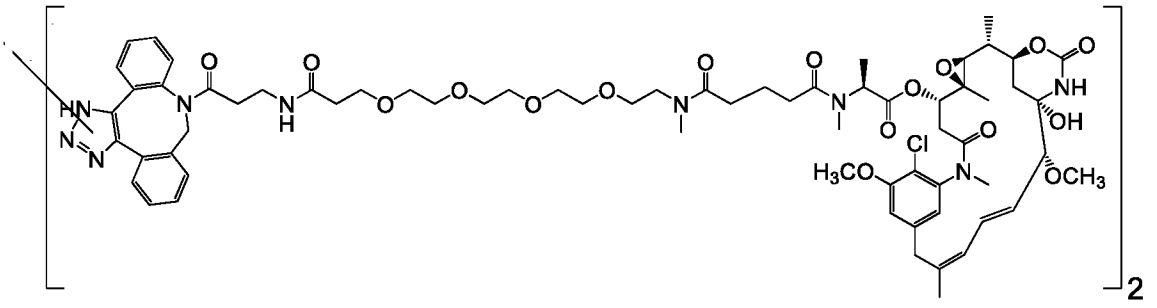
[00192] In some embodiments, provided herein are anti-FOLR1 conjugates comprising a modified hemiasterlin and linker as described, for example, in PCT Publication No.

WO 2016/123582. For example, the conjugate can have a structure comprising any of Formulas 1000-1000b, 1001-1001b, 1002-1002b, and I-XIXb-2, 101-111b, or 1-8b as described in PCT Publication No. WO 2016/2016/123582. Examples of conjugates comprising a modified hemiasterlin and linker are provided below.

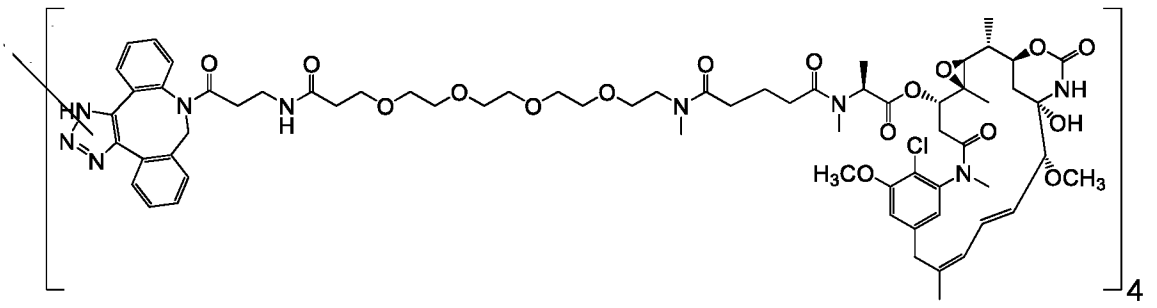
[00193] In some embodiments, provided herein are anti-FOLR1 conjugates having the structure of **Conjugate M**:



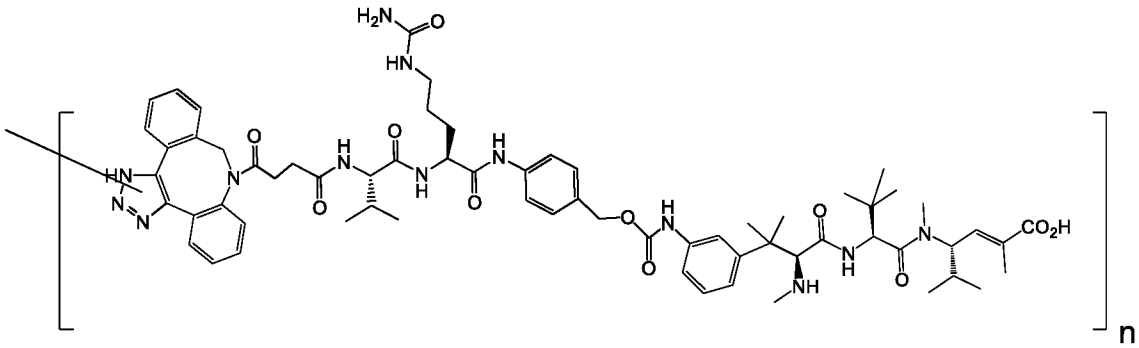
where **n** is an integer from 1 to 6. In some embodiments, **n** is an integer from 1 to 4. In some embodiments, **n** is 2. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



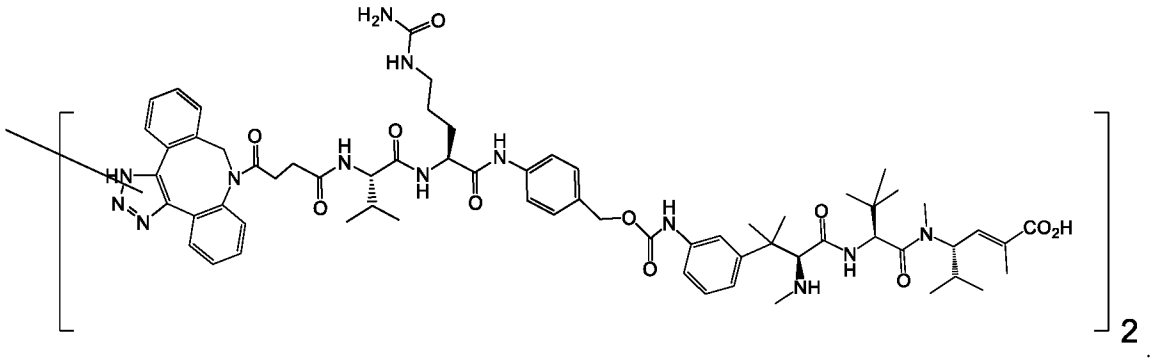
In some embodiments, **n** is 4. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



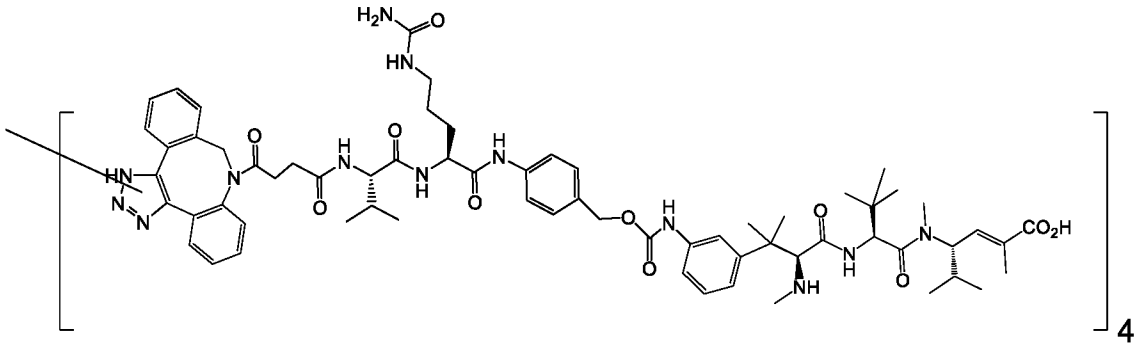
[00194] In some embodiments, provided herein are anti-FOLR1 conjugates having the structure of **Conjugate P**:



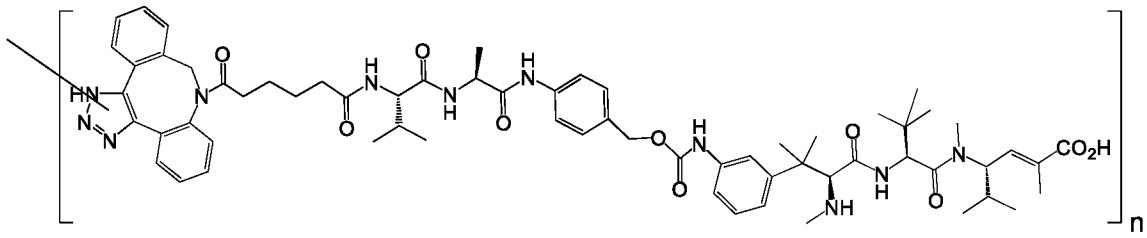
where **n** is an integer from 1 to 6. In some embodiments, **n** is an integer from 1 to 4. In some embodiments, **n** is 2. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



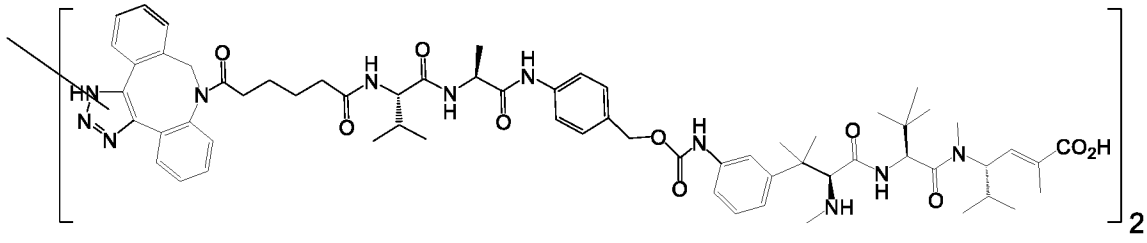
In some embodiments, **n** is 4. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



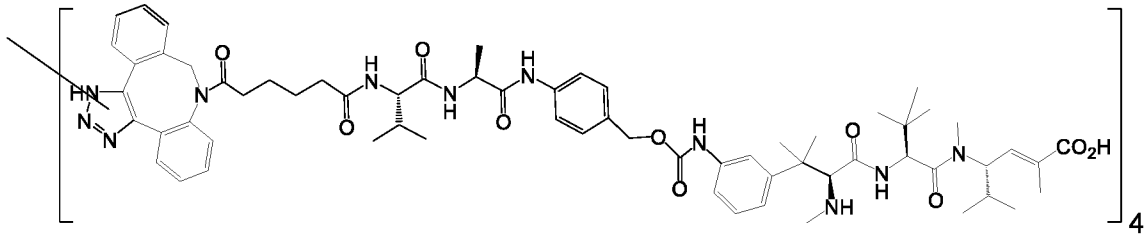
[00195] In some embodiments, provided herein are anti-FOLR1 conjugates having the structure of **Conjugate Q**:



where **n** is an integer from 1 to 6. In some embodiments, **n** is an integer from 1 to 4. In some embodiments, **n** is 2. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



In some embodiments, **n** is 4. For example, in some embodiments, the anti-FOLR1 conjugate has the structure:



[00196] In any of the foregoing embodiments wherein the anti-FOLR1 conjugate has a structure according to **Conjugate M**, **Conjugate P**, or **Conjugate Q**, the bracketed structure can be covalently bonded to one or more non-natural amino acids of the antibody, wherein the one or more non-natural amino acids are located at sites selected from the group consisting of: HC-F404, HC-Y180, and LC-K42 according to the Kabat or EU numbering scheme of Kabat. In some embodiments, the bracketed structure is covalently bonded to one or more non-natural amino acids at site HC-F404 of the antibody. In some embodiments, the bracketed structure is covalently bonded to one or more non-natural amino acids at site HC-Y180 of the antibody. In some embodiments, the bracketed structure is covalently bonded to one or more non-natural amino acids at site LC-K42 of the antibody. In some embodiments, the bracketed structure is covalently bonded to one or more non-natural amino acids at sites HC-F404 and HC-Y180 of the antibody. In some embodiments, at least one bracketed structure is covalently bonded to a

non-natural amino acid at site HC-F404 of the antibody, and at least one bracketed structure is covalently bonded a non-natural amino acid at site HC-Y180 of the antibody. In some embodiments, the bracketed structure is covalently bonded to one or more non-natural amino acids at sites HC-Y180 and LC-K42 of the antibody. In some embodiments, at least one bracketed structure is covalently bonded to a non-natural amino acid at site HC-Y180 of the antibody, and at least one bracketed structure is covalently bonded a non-natural amino acid at site LC-K32 of the antibody.

3. Payloads

[00197] In addition to the payloads described above, the molecular payload can be any molecular entity that one of skill in the art might desire to conjugate to the polypeptide. In certain embodiments, the payload is a therapeutic moiety. In such embodiment, the antibody conjugate can be used to target the therapeutic moiety to its molecular target. In certain embodiments, the payload is a labeling moiety. In such embodiments, the antibody conjugate can be used to detect binding of the polypeptide to its target. In certain embodiments, the payload is a cytotoxic moiety. In such embodiments, the antibody conjugate can be used target the cytotoxic moiety to a diseased cell, for example a cancer cell, to initiate destruction or elimination of the cell. Conjugates comprising other molecular payloads apparent to those of skill in the art are within the scope of the conjugates described herein.

[00198] In certain embodiments, an antibody conjugate can have a payload selected from the group consisting of a label, a dye, a polymer, a water-soluble polymer, polyethylene glycol, a derivative of polyethylene glycol, a photocrosslinker, a cytotoxic compound, a radionuclide, a drug, an affinity label, a photoaffinity label, a reactive compound, a resin, a second protein or polypeptide or polypeptide analog, an antibody or antibody fragment, a metal chelator, a cofactor, a fatty acid, a carbohydrate, a polynucleotide, a DNA, a RNA, an antisense polynucleotide, a peptide, a water-soluble dendrimer, a cyclodextrin, an inhibitory ribonucleic acid, a biomaterial, a nanoparticle, a spin label, a fluorophore, a metal-containing moiety, a radioactive moiety, a novel functional group, a group that covalently or noncovalently interacts with other molecules, a photocaged moiety, a photoisomerizable moiety, biotin, a derivative of biotin, a biotin analogue, a moiety incorporating a heavy atom, a chemically cleavable group, a photocleavable group, an elongated side chain, a carbon-linked sugar, a redox-active agent, an amino thioacid, a toxic moiety, an isotopically labeled moiety, a biophysical probe, a phosphorescent group, a chemiluminescent group, an electron dense group, a magnetic group,

an intercalating group, a chromophore, an energy transfer agent, a biologically active agent, a detectable label, a small molecule, or any combination thereof. In an embodiment, the payload is a label, a dye, a polymer, a cytotoxic compound, a radionuclide, a drug, an affinity label, a resin, a protein, a polypeptide, a polypeptide analog, an antibody, antibody fragment, a metal chelator, a cofactor, a fatty acid, a carbohydrate, a polynucleotide, a DNA, a RNA, a peptide, a fluorophore, or a carbon-linked sugar. In another embodiment, the payload is a label, a dye, a polymer, a drug, an antibody, antibody fragment, a DNA, an RNA, or a peptide.

[00199] Useful drug payloads include any cytotoxic, cytostatic or immunomodulatory agent. Useful classes of cytotoxic or immunomodulatory agents include, for example, antitubulin agents, auristatins, DNA minor groove binders, DNA replication inhibitors, alkylating agents (*e.g.*, platinum complexes such as cis-platin, mono(platinum), bis(platinum) and tri-nuclear platinum complexes and carboplatin), anthracyclines, antibiotics, antifolates, antimetabolites, calmodulin inhibitors, chemotherapy sensitizers, duocarmycins, etoposides, fluorinated pyrimidines, ionophores, lexitropsins, maytansinoids, nitrosoureas, platinols, pore-forming compounds, purine antimetabolites, puromycins, radiation sensitizers, rapamycins, steroids, taxanes, topoisomerase inhibitors, vinca alkaloids, or the like.

[00200] Individual cytotoxic or immunomodulatory agents include, for example, an androgen, anthramycin (AMC), asparaginase, 5-azacytidine, azathioprine, bleomycin, busulfan, buthionine sulfoximine, calicheamicin, calicheamicin derivatives, camptothecin, carboplatin, carmustine (BSNU), CC-1065, chlorambucil, cisplatin, colchicine, cyclophosphamide, cytarabine, cytidine arabinoside, cytochalasin B, dacarbazine, dactinomycin (formerly actinomycin), daunorubicin, decarbazine, DM1, DM4, docetaxel, doxorubicin, etoposide, an estrogen, 5-fluordeoxyuridine, 5-fluorouracil, gemcitabine, gramicidin D, hydroxyurea, idarubicin, ifosfamide, irinotecan, lomustine (CCNU), maytansine, mechlorethamine, melphalan, 6-mercaptopurine, methotrexate, mithramycin, mitomycin C, mitoxantrone, nitroimidazole, paclitaxel, palytoxin, plicamycin, procarbizine, rhizoxin, streptozotocin, tenoposide, 6-thioguanine, thioTEPA, topotecan, vinblastine, vincristine, vinorelbine, VP-16 and VM-26.

[00201] In some embodiments, suitable cytotoxic agents include, for example, DNA minor groove binders (*e.g.*, enediynes and lexitropsins, a CBI compound; see also U.S. Pat. No. 6,130,237), duocarmycins, taxanes (*e.g.*, paclitaxel and docetaxel), puromycins, vinca alkaloids, CC-1065, SN-38, topotecan, morpholino-doxorubicin, rhizoxin, cyanomorpholino-

doxorubicin, echinomycin, combretastatin, netropsin, epothilone A and B, estramustine, cryptophycins, cemadotin, maytansinoids, discodermolide, eleutherobin, and mitoxantrone.

[00202] In some embodiments, the payload is an anti-tubulin agent. Examples of anti-tubulin agents include, but are not limited to, taxanes (*e.g.*, Taxol® (paclitaxel), Taxotere® (docetaxel)), T67 (Tularik) and vinca alkyloids (*e.g.*, vincristine, vinblastine, vindesine, and vinorelbine). Other antitubulin agents include, for example, baccatin derivatives, taxane analogs, epothilones (*e.g.*, epothilone A and B), nocodazole, colchicine and colcimid, estramustine, cryptophycins, cemadotin, maytansinoids, combretastatins, discodermolide, and eleutherobin.

[00203] In certain embodiments, the cytotoxic agent is a maytansinoid, another group of anti-tubulin agents. For example, in specific embodiments, the maytansinoid can be maytansine or DM-1 (ImmunoGen, Inc.; see also Chari *et al.*, 1992, Cancer Res. 52:127-131).

[00204] In some embodiments, the payload is an auristatin, such as auristatin E or a derivative thereof. For example, the auristatin E derivative can be an ester formed between auristatin E and a keto acid. For example, auristatin E can be reacted with paraacetyl benzoic acid or benzoylvaleric acid to produce AEB and AEVB, respectively. Other typical auristatin derivatives include AFP (auristatin phenylalanine phenylenediamine), MMAF (monomethyl auristatin F), and MMAE (monomethyl auristatin E). The synthesis and structure of auristatin derivatives are described in U.S. Patent Application Publication Nos. 2003-0083263, 2005-0238649 and 2005-0009751; International Patent Publication No. WO 04/010957, International Patent Publication No. WO 02/088172, and U.S. Pat. Nos. 6,323,315; 6,239,104; 6,034,065; 5,780,588; 5,665,860; 5,663,149; 5,635,483; 5,599,902; 5,554,725; 5,530,097; 5,521,284; 5,504,191; 5,410,024; 5,138,036; 5,076,973; 4,986,988; 4,978,744; 4,879,278; 4,816,444; and 4,486,414.

[00205] In some embodiments, the payload is a hemiasterlin. Hemiasterlins suitable for use in the antibody-drug conjugates described herein are described, for example, in International Patent Publication No. WO 2016/2016/123582, which is incorporated herein by reference in its entirety.

[00206] In some embodiments, the payload is not a radioisotope. In some embodiments, the payload is not radioactive.

[00207] In some embodiments, the payload is an antimetabolite. The antimetabolite can be, for example, a purine antagonist (*e.g.*, azothioprine or mycophenolate mofetil), a dihydrofolate

reductase inhibitor (*e.g.*, methotrexate), acyclovir, gangcyclovir, zidovudine, vidarabine, ribavarin, azidothymidine, cytidine arabinoside, amantadine, dideoxyuridine, iododeoxyuridine, poscarnet, or trifluridine.

[00208] In other embodiments, the payload is tacrolimus, cyclosporine, FU506 or rapamycin. In further embodiments, the Drug is aldesleukin, alemtuzumab, alitretinoin, allopurinol, altretamine, amifostine, anastrozole, arsenic trioxide, bexarotene, bexarotene, calusterone, capecitabine, celecoxib, cladribine, Darbepoetin alfa, Denileukin diftitox, dexrazoxane, dromostanolone propionate, epirubicin, Epoetin alfa, estramustine, exemestane, Filgrastim, floxuridine, fludarabine, fulvestrant, gemcitabine, gemtuzumab ozogamicin (MYLOTARG), goserelin, idarubicin, ifosfamide, imatinib mesylate, Interferon alfa-2a, irinotecan, letrozole, leucovorin, levamisole, meclorethamine or nitrogen mustard, megestrol, mesna, methotrexate, methoxsalen, mitomycin C, mitotane, nandrolone phenpropionate, oprelvekin, oxaliplatin, pamidronate, pegademase, pegaspargase, pegfilgrastim, pentostatin, pipobroman, a pladienolide, plicamycin, porfimer sodium, procarbazine, quinacrine, rasburicase, Rituximab, Sargramostim, streptozocin, tamoxifen, temozolomide, teniposide, testolactone, thioguanine, toremifene, Tositumomab, Trastuzumab (HERCEPTIN), tretinoin, uracil mustard, valrubicin, vinblastine, vincristine, vinorelbine or zoledronate.

[00209] In some embodiments, the payload is an immunomodulatory agent. The immunomodulatory agent can be, for example, gangcyclovir, etanercept, tacrolimus, cyclosporine, rapamycin, cyclophosphamide, azathioprine, mycophenolate mofetil or methotrexate. Alternatively, the immunomodulatory agent can be, for example, a glucocorticoid (*e.g.*, cortisol or aldosterone) or a glucocorticoid analogue (*e.g.*, prednisone or dexamethasone).

[00210] In some embodiments, the immunomodulatory agent is an agent that modulates components of the immune system such that it would enhance the anti-tumor activity of the conjugate. Such agents would include but are not limited to agonists of Toll-Like receptors (*e.g.*, poly-ICLC (Hiltonol), GLA, MEDI9197, VTX-2337 (Motolimid), CpG (SD-101), and IMO-2125); agonists of the STING (stimulator of interferon genes) pathway (*e.g.*, MK-1454, ADU-S100, and SB11285); activators of RIG-I-Like Receptor (RLR) signaling (*e.g.*, RGT100); inhibitors of IDO-1 (indoleamine 2,3-dioxygenase 1) (*e.g.*, GDC-0919 (navoximod), BMS-986205, and epacadostat); and small molecule blockers of the PD-1 pathway (*e.g.*, CA-170, BMS-8, BMS-202, and AUNP12).

[00211] In some embodiments, the immunomodulatory agent is an anti-inflammatory agent, such as arylcarboxylic derivatives, pyrazole-containing derivatives, oxicam derivatives and nicotinic acid derivatives. Classes of anti-inflammatory agents include, for example, cyclooxygenase inhibitors, 5-lipoxygenase inhibitors, and leukotriene receptor antagonists. In some embodiments, the immunomodulatory agent is a cytokine, such as, *e.g.*, IL-12.

[00212] Suitable cyclooxygenase inhibitors include meclofenamic acid, mefenamic acid, carprofen, diclofenac, diflunisal, fenbufen, fenoprofen, indomethacin, ketoprofen, nabumetone, sulindac, tenoxicam and tolmetin.

[00213] Suitable lipoxygenase inhibitors include redox inhibitors (*e.g.*, catechol butane derivatives, nordihydroguaiaretic acid (NDGA), masoprocol, phenidone, Ianopalen, indazolinones, naphazatrom, benzofuranol, alkylhydroxylamine), and non-redox inhibitors (*e.g.*, hydroxythiazoles, methoxyalkylthiazoles, benzopyrans and derivatives thereof, methoxytetrahydropyran, boswellic acids and acetylated derivatives of boswellic acids, and quinolinemethoxyphenylacetic acids substituted with cycloalkyl radicals), and precursors of redox inhibitors.

[00214] Other suitable lipoxygenase inhibitors include antioxidants (*e.g.*, phenols, propyl gallate, flavonoids and/or naturally occurring substrates containing flavonoids, hydroxylated derivatives of the flavones, flavonol, dihydroquercetin, luteolin, galangin, orobol, derivatives of chalcone, 4,2',4'-trihydroxychalcone, ortho-aminophenols, N-hydroxyureas, benzofuranols, ebselen and species that increase the activity of the reducing selenoenzymes), iron chelating agents (*e.g.*, hydroxamic acids and derivatives thereof, N-hydroxyureas, 2-benzyl-1-naphthol, catechols, hydroxylamines, carnosol trolox C, catechol, naphthol, sulfasalazine, zyleuton, 5-hydroxyanthranilic acid and 4-(omega-arylalkyl)phenylalkanoic acids), imidazole-containing compounds (*e.g.*, ketoconazole and itraconazole), phenothiazines, and benzopyran derivatives.

[00215] Yet other suitable lipoxygenase inhibitors include inhibitors of eicosanoids (*e.g.*, octadecatetraenoic, eicosatetraenoic, docosapentaenoic, eicosahexaenoic and docosahexaenoic acids and esters thereof, PGE1 (prostaglandin E1), PGA2 (prostaglandin A2), viprostol, 15-monohydroxyeicosatetraenoic, 15-monohydroxy-eicosatrienoic and 15-monohydroxyeicosapentaenoic acids, and leukotrienes B5, C5 and D5), compounds interfering with calcium flows, phenothiazines, diphenylbutylamines, verapamil, fuscoside, curcumin, chlorogenic acid, caffeic acid, 5,8,11,14-eicosatetrayenoic acid (ETYA),

hydroxyphenylretinamide, Ionapalen, esculin, diethylcarbamazine, phenantroline, baicalein, proxicromil, thioethers, diallyl sulfide and di-(1-propenyl) sulfide.

[00216] Leukotriene receptor antagonists include calcitriol, ontazolast, Bayer Bay-x-1005, Ciba-Geigy CGS-25019C, ebselen, Leo Denmark ETH-615, Lilly LY-293111, Ono ONO-4057, Terumo TMK-688, Boehringer Ingleheim BI-RM-270, Lilly LY 213024, Lilly LY 264086, Lilly LY 292728, Ono ONO LB457, Pfizer 105696, Perdue Frederick PF 10042, Rhone-Poulenc Rorer RP 66153, SmithKline Beecham SB-201146, SmithKline Beecham SB-201993, SmithKline Beecham SB-209247, Searle SC-53228, Sumitamo SM 15178, American Home Products WAY 121006, Bayer Bay-o-8276, Warner-Lambert CI-987, Warner-Lambert CI-987BPC-15LY 223982, Lilly LY 233569, Lilly LY-255283, MacroNex MNX-160, Merck and Co. MK-591, Merck and Co. MK-886, Ono ONO-LB-448, Purdue Frederick PF-5901, Rhone-Poulenc Rorer RG14893, Rhone-Poulenc Rorer RP 66364, Rhone-Poulenc Rorer RP 69698, Shionoogi S-2474, Searle SC-41930, Searle SC-50505, Searle SC-51146, Searle SC-52798, SmithKline Beecham SK&F-104493, Leo Denmark SR-2566, Tanabe T-757 and Teijin TEI-1338.

[00217] Other useful drug payloads include chemical compounds useful in the treatment of cancer. Examples of chemotherapeutic agents include Erlotinib (TARCEVA®, Genentech/OSI Pharm.), Bortezomib (VELCADE®, Millennium Pharm.), Fulvestrant (FASLODEX®, AstraZeneca), Sutent (SU11248, Pfizer), Letrozole (FEMARA®, Novartis), Imatinib mesylate (GLEEVEC®, Novartis), PTK787/ZK 222584 (Novartis), Oxaliplatin (Eloxatin®, Sanofi), 5-FU (5-fluorouracil), Leucovorin, Rapamycin (Sirolimus, RAPAMUNE®, Wyeth), Lapatinib (TYKERB®, GSK572016, Glaxo Smith Kline), Lonafarnib (SCH 66336), Sorafenib (BAY43-9006, Bayer Labs), and Gefitinib (IRESSA®, AstraZeneca), AG1478, AG1571 (SU 5271; Sugen), alkylating agents such as thiotepa and CYTOXAN® cyclosphosphamide; alkyl sulfonates such as busulfan, improsulfan and piposulfan; aziridines such as benzodopa, carboquone, meturedopa, and uredopa; ethylenimines and methylamelamines including altretamine, triethylenemelamine, triethylenephosphoramide, triethylenethiophosphoramide and trimethylomelamine; acetogenins (especially bullatacin and bullatacinone); a camptothecin (including the synthetic analog topotecan); bryostatin; callystatin; CC-1065 (including its adozelesin, carzelesin and bizelesin synthetic analogs); cryptophycins (particularly cryptophycin 1 and cryptophycin 8); dolastatin; duocarmycin (including the synthetic analogs, KW-2189 and CB1-TM1); eleutherobin; pancratistatin; a sarcodictyin; spongistatin; nitrogen mustards such as chlorambucil, chlomaphazine, chlorophosphamide, estramustine, ifosfamide,

mechlorethamine, mechlorethamine oxide hydrochloride, melphalan, novembichin, phenesterine, prednimustine, trofosfamide, uracil mustard; nitrosureas such as carmustine, chlorozotocin, fotemustine, lomustine, nimustine, and ranimnustine; antibiotics such as the enediyne antibiotics (*e.g.*, calicheamicin, especially uncialamycin, calicheamicin gammall, and calicheamicin omegall (Angew Chem. Intl. Ed. Engl. (1994) 33:183-186); dynemicin, including dynemicin A; bisphosphonates, such as clodronate; an esperamicin; as well as neocarzinostatin chromophore and related chromoprotein enediyne antibiotic chromophores), aclacinomysins, actinomycin, authramycin, azaserine, bleomycins, cactinomycin, carabicin, caminomycin, carzinophilin, chromomycinis, dactinomycin, daunorubicin, detorubicin, 6-diazo-5-oxo-L-norleucine, ADRIAMYCIN® (doxorubicin), morpholino-doxorubicin, cyanomorpholino-doxorubicin, 2-pyrrolino-doxorubicin and deoxydoxorubicin), epirubicin, esorubicin, idarubicin, marcellomycin, mitomycins such as mitomycin C, mycophenolic acid, nogalamycin, olivomycins, peplomycin, porfiromycin, puromycin, quelamycin, rodorubicin, streptonigrin, streptozocin, tubercidin, ubenimex, zinostatin, zorubicin; anti-metabolites such as methotrexate and 5-fluorouracil (5-FU); folic acid analogs such as denopterin, methotrexate, pladienolide B, pteropterin, trimetrexate; purine analogs such as fludarabine, 6-mercaptopurine, thiamniprine, thioguanine; pyrimidine analogs such as ancitabine, azacitidine, 6-azauridine, carmofur, cytarabine, dideoxyuridine, doxifluridine, enocitabine, floxuridine; androgens such as calusterone, dromostanolone propionate, epitiostanol, mepitiostane, testolactone; anti-adrenals such as aminoglutethimide, mitotane, trilostane; folic acid replenisher such as frolinic acid; aceglatone; aldophosphamide glycoside; aminolevulinic acid; eniluracil; amsacrine; bestrabucil; bisantrene; edatraxate; defofamine; demecolcine; diaziquone; elformithine; elliptinium acetate; an epothilone; etoglucid; gallium nitrate; hydroxyurea; lentinan; lonidainine; maytansinoids such as maytansine and ansamitocins; mitoguazone; mitoxantrone; mopidanmol; nitraerine; pentostatin; phenamet; pirarubicin; losoxantrone; podophyllinic acid; 2-ethylhydrazide; procarbazine; PSK® polysaccharide complex (JHS Natural Products, Eugene, Oreg.); razoxane; rhizoxin; sizofuran; spirogermanium; tenuazonic acid; triaziquone; 2,2',2''-trichlorotriethylamine; trichothecenes (especially T-2 toxin, verracurin A, roridin A and anguidine); urethan; vindesine; dacarbazine; mannomustine; mitobronitol; mitolactol; pipobroman; gacytosine; arabinoside ("Ara-C"); cyclophosphamide; thiotepa; taxoids, *e.g.*, TAXOL® (paclitaxel; Bristol-Myers Squibb Oncology, Princeton, N.J.), ABRAXANE® (Cremophor-free), albumin-engineered nanoparticle formulations of paclitaxel (American Pharmaceutical Partners, Schaumberg, Ill.),

and TAXOTERE® (doxetaxel; Rhone-Poulenc Rorer, Antony, France); chloranmbucil; GEMZAR® (gemcitabine); 6-thioguanine; mercaptopurine; methotrexate; platinum analogs such as cisplatin and carboplatin; vinblastine; etoposide (VP-16); ifosfamide; mitoxantrone; vincristine; NAVELBINE® (vinorelbine); novantrone; teniposide; edatrexate; daunomycin; aminopterin; capecitabine (XELODA®); ibandronate; CPT-11; topoisomerase inhibitor RFS 2000; difluoromethylornithine (DMFO); retinoids such as retinoic acid; and pharmaceutically acceptable salts, acids and derivatives of any of the above.

[00218] Other useful payloads include: (i) anti-hormonal agents that act to regulate or inhibit hormone action on tumors such as anti-estrogens and selective estrogen receptor modulators (SERMs), including, for example, tamoxifen (including NOLVADEX®; tamoxifen citrate), raloxifene, droloxifene, 4-hydroxytamoxifen, trioxifene, keoxifene, LY117018, onapristone, and FARESTON® (toremifine citrate); (ii) aromatase inhibitors that inhibit the enzyme aromatase, which regulates estrogen production in the adrenal glands, such as, for example, 4(5)-imidazoles, aminoglutethimide, MEGASE® (megestrol acetate), AROMASIN® (exemestane; Pfizer), formestanie, fadrozole, RIVISOR® (vorozole), FEMARA® (letrozole; Novartis), and ARIMIDEX® (anastrozole; AstraZeneca); (iii) anti-androgens such as flutamide, nilutamide, bicalutamide, leuprolide, and goserelin; as well as troxacitabine (a 1,3-dioxolane nucleoside cytosine analog); (iv) protein kinase inhibitors; (v) lipid kinase inhibitors; (vi) antisense oligonucleotides, particularly those which inhibit expression of genes in signaling pathways implicated in aberrant cell proliferation, such as, for example, PKC-alpha, Ralf and H-Ras; (vii) ribozymes such as VEGF expression inhibitors (*e.g.*, ANGIOZYME®) and HER2 expression inhibitors; (viii) vaccines such as gene therapy vaccines, for example, ALLOVECTIN®, LEUVECTIN®, and VAXID®; PROLEUKIN® rIL-2; a topoisomerase 1 inhibitor such as LURTOTECAN®; ABARELIX® rmRH; (ix) anti-angiogenic agents such as bevacizumab (AVASTIN®, Genentech); (x) agents that act to regulate or inhibit activity of members of the poly(ADP-ribose) polymerase (PARP) family in tumors (*e.g.*, Talazoparib (BMN-673), Iniparib (BSI 201), Veliparib (ABT-888), Olaparib (AZD-2281, trade name **LYNPARZA™**), Rucaparib (AG 014699), BGB-290, E7016, E7449, and CEP-9722); (xi) agents that act to regulate or inhibit activity of members of the histone deacetylase (HDAC) family in tumors (*e.g.*, abexinostat, entinostat, gavinostat, 4SC-202, ACY-241, AR-42, CG200745, CHR-2845, CHR-3996, CXD101, MPT0E028, OBP-801, SHP-141, CUDC-101, KA2507, panobinostat, pracinostat, quisinostat, resminostat, ricolinostat); (xii) agents that act to regulate or inhibit activity of mitochondrial enzyme isocitrate dehydrogenase type 2

(IDH2) in tumors (*e.g.*, enasidenib mesylate (CC-90007, AG-221 mesylate); and (xiii) pharmaceutically acceptable salts, acids and derivatives of any of the above. Other useful payloads include anti-angiogenic agents, including, *e.g.*, MMP-2 (matrix-metalloproteinase 2) inhibitors, MMP-9 (matrix-metalloproteinase 9) inhibitors, COX-II (cyclooxygenase II) inhibitors, and VEGF receptor tyrosine kinase inhibitors. Examples of such useful matrix metalloproteinase inhibitors that can be used in combination with the present compounds/compositions are described in WO 96/33172, WO 96/27583, EP 818442, EP 1004578, WO 98/07697, WO 98/03516, WO 98/34918, WO 98/34915, WO 98/33768, WO 98/30566, EP 606,046, EP 931,788, WO 90/05719, WO 99/52910, WO 99/52889, WO 99/29667, WO 99/07675, EP 945864, U.S. Pat. No. 5,863,949, U.S. Pat. No. 5,861,510, and EP 780,386, all of which are incorporated herein in their entireties by reference. Examples of VEGF receptor tyrosine kinase inhibitors include 4-(4-bromo-2-fluoroanilino)-6-methoxy-7-(1-methylpiperidin-4-ylmethoxy)quinazoline (ZD6474; Example 2 within WO 01/32651), 4-(4-fluoro-2-methylindol-5-yloxy)-6-methoxy-7-(3-pyrrolidin-1-ylpropoxy)quinazoline (AZD2171; Example 240 within WO 00/47212), vatalanib (PTK787; WO 98/35985) and SU11248 (sunitinib; WO 01/60814), and compounds such as those disclosed in PCT Publication Nos. WO 97/22596, WO 97/30035, WO 97/32856, and WO 98/13354).

[00219] In certain embodiments, the payload is an antibody or an antibody fragment. In certain embodiments, the payload antibody or fragment can be encoded by any of the immunoglobulin genes recognized by those of skill in the art. The immunoglobulin genes include, but are not limited to, the κ, λ, α, γ (IgG1, IgG2, IgG3, and IgG4), δ, ε and μ constant region genes, as well as the immunoglobulin variable region genes. The term includes full-length antibody and antibody fragments recognized by those of skill in the art, and variants thereof. Exemplary fragments include but are not limited to Fv, Fc, Fab, and $(Fab')_2$, single chain Fv (scFv), diabodies, triabodies, tetrabodies, bifunctional hybrid polypeptides, CDR1, CDR2, CDR3, combinations of CDR's, variable regions, framework regions, constant regions, and the like.

[00220] In certain embodiments, the payload is one or more water-soluble polymers. A wide variety of macromolecular polymers and other molecules can be linked to the polypeptides described herein to modulate biological properties of the polypeptide, and/or provide new biological properties to the polypeptide. These macromolecular polymers can be linked to the polypeptide via a naturally encoded amino acid, via a non-naturally encoded amino acid, or any functional substituent of a natural or modified amino acid, or any substituent or functional

group added to a natural or modified amino acid. The molecular weight of the polymer may be of a wide range, including but not limited to, between about 100 Da and about 100,000 Da or more.

[00221] The polymer selected may be water soluble so that a protein to which it is attached does not precipitate in an aqueous environment, such as a physiological environment. The polymer may be branched or unbranched. Preferably, for therapeutic use of the end-product preparation, the polymer will be pharmaceutically acceptable.

[00222] In certain embodiments, the proportion of polyethylene glycol molecules to polypeptide molecules will vary, as will their concentrations in the reaction mixture. In general, the optimum ratio (in terms of efficiency of reaction in that there is minimal excess unreacted protein or polymer) may be determined by the molecular weight of the polyethylene glycol selected and on the number of available reactive groups available. As relates to molecular weight, typically the higher the molecular weight of the polymer, the fewer number of polymer molecules which may be attached to the protein. Similarly, branching of the polymer should be taken into account when optimizing these parameters. Generally, the higher the molecular weight (or the more branches) the higher the polymer:protein ratio.

[00223] The water soluble polymer may be any structural form including but not limited to linear, forked or branched. Typically, the water soluble polymer is a poly(alkylene glycol), such as poly(ethylene glycol) (PEG), but other water soluble polymers can also be employed. By way of example, PEG is used to describe certain embodiments.

[00224] PEG is a well-known, water soluble polymer that is commercially available or can be prepared by ring-opening polymerization of ethylene glycol according to methods well known in the art (Sandler and Karo, Polymer Synthesis, Academic Press, New York, Vol. 3, pages 138-161). The term "PEG" is used broadly to encompass any polyethylene glycol molecule, without regard to size or to modification at an end of the PEG, and can be represented as linked to a polypeptide by the formula: $XO–(CH_2CH_2O)_n–CH_2CH_2–Y$ where n is 2 to 10,000,X is H or a terminal modification, including but not limited to, a C_{1-4} alkyl, and Y is the attachment point to the polypeptide.

[00225] In some cases, a PEG terminates on one end with hydroxy or methoxy, i.e., X is H or CH_3 ("methoxy PEG"). Alternatively, the PEG can terminate with a reactive group, thereby forming a bifunctional polymer. Typical reactive groups can include those reactive groups that are commonly used to react with the functional groups found in the 20 common amino acids

(including but not limited to, maleimide groups, activated carbonates (including but not limited to, p-nitrophenyl ester), activated esters (including but not limited to, N-hydroxysuccinimide, p-nitrophenyl ester, and aldehydes) as well as functional groups that are inert to the 20 common amino acids but that react specifically with complementary functional groups present in non-naturally encoded amino acids (including but not limited to, azide groups, alkyne groups). It is noted that the other end of the PEG, which is shown in the above formula by Y, will attach either directly or indirectly to a polypeptide via a naturally-occurring or non-naturally encoded amino acid. For instance, Y may be an amide, carbamate or urea linkage to an amine group (including but not limited to, the epsilon amine of lysine or the N-terminus) of the polypeptide. Alternatively, Y may be a maleimide linkage to a thiol group (including but not limited to, the thiol group of cysteine). Alternatively, Y may be a linkage to a residue not commonly accessible via the 20 common amino acids. For example, an azide group on the PEG can be reacted with an alkyne group on the polypeptide to form a Huisgen [3+2] cycloaddition product. Alternatively, an alkyne group on the PEG can be reacted with an azide group present in a non-naturally encoded amino acid, such as the modified amino acids described herein, to form a similar product. In some embodiments, a strong nucleophile (including but not limited to, hydrazine, hydrazide, hydroxylamine, semicarbazide) can be reacted with an aldehyde or ketone group present in a non-naturally encoded amino acid to form a hydrazone, oxime or semicarbazone, as applicable, which in some cases can be further reduced by treatment with an appropriate reducing agent. Alternatively, the strong nucleophile can be incorporated into the polypeptide via a non-naturally encoded amino acid and used to react preferentially with a ketone or aldehyde group present in the water soluble polymer.

[00226] Any molecular mass for a PEG can be used as practically desired, including but not limited to, from about 100 Daltons (Da) to 100,000 Da or more as desired (including but not limited to, sometimes 0.1-50 kDa or 10-40 kDa). Branched chain PEGs, including but not limited to, PEG molecules with each chain having a MW ranging from 1-100 kDa (including but not limited to, 1-50 kDa or 5-20 kDa) can also be used. A wide range of PEG molecules are described in, including but not limited to, the Shearwater Polymers, Inc. catalog, and the Nektar Therapeutics catalog, incorporated herein by reference.

[00227] Generally, at least one terminus of the PEG molecule is available for reaction with the antibody. For example, PEG derivatives bearing alkyne and azide moieties for reaction with amino acid side chains can be used to attach PEG to non-naturally encoded amino acids as described herein. If the non-naturally encoded amino acid comprises an azide, then the PEG

will typically contain either an alkyne moiety to effect formation of the [3+2] cycloaddition product or an activated PEG species (i.e., ester, carbonate) containing a phosphine group to effect formation of the amide linkage. Alternatively, if the non-naturally encoded amino acid comprises an alkyne, then the PEG will typically contain an azide moiety to effect formation of the [3+2] Huisgen cycloaddition product. If the non-naturally encoded amino acid comprises a carbonyl group, the PEG will typically comprise a potent nucleophile (including but not limited to, a hydrazide, hydrazine, hydroxylamine, or semicarbazide functionality) in order to effect formation of corresponding hydrazone, oxime, and semicarbazone linkages, respectively. In other alternatives, a reverse of the orientation of the reactive groups described herein can be used, i.e., an azide moiety in the non-naturally encoded amino acid can be reacted with a PEG derivative containing an alkyne.

[00228] In some embodiments, the polypeptide variant with a PEG derivative contains a chemical functionality that is reactive with the chemical functionality present on the side chain of the non-naturally encoded amino acid.

[00229] In certain embodiments, the payload is an azide- or acetylene-containing polymer comprising a water soluble polymer backbone having an average molecular weight from about 800 Da to about 100,000 Da. The polymer backbone of the water-soluble polymer can be poly(ethylene glycol). However, it should be understood that a wide variety of water soluble polymers including but not limited to poly(ethylene)glycol and other related polymers, including poly(dextran) and poly(propylene glycol), are also suitable for use and that the use of the term PEG or poly(ethylene glycol) is intended to encompass and include all such molecules. The term PEG includes, but is not limited to, poly(ethylene glycol) in any of its forms, including bifunctional PEG, multiarmed PEG, derivatized PEG, forked PEG, branched PEG, pendent PEG (i.e. PEG or related polymers having one or more functional groups pendent to the polymer backbone), or PEG with degradable linkages therein.

[00230] The polymer backbone can be linear or branched. Branched polymer backbones are generally known in the art. Typically, a branched polymer has a central branch core moiety and a plurality of linear polymer chains linked to the central branch core. PEG is commonly used in branched forms that can be prepared by addition of ethylene oxide to various polyols, such as glycerol, glycerol oligomers, pentaerythritol and sorbitol. The central branch moiety can also be derived from several amino acids, such as lysine. The branched poly(ethylene glycol) can be represented in general form as $R(\text{-PEG-OH})_m$ in which R is derived from a core moiety,

such as glycerol, glycerol oligomers, or pentaerythritol, and m represents the number of arms. Multi-armed PEG molecules, such as those described in U.S. Pat. Nos. 5,932,462 5,643,575; 5,229,490; 4,289,872; U.S. Pat. Appl. 2003/0143596; WO 96/21469; and WO 93/21259, each of which is incorporated by reference herein in its entirety, can also be used as the polymer backbone.

[00231] Branched PEG can also be in the form of a forked PEG represented by $PEG(-YCHZ_2)_n$, where Y is a linking group and Z is an activated terminal group linked to CH by a chain of atoms of defined length.

[00232] Yet another branched form, the pendant PEG, has reactive groups, such as carboxyl, along the PEG backbone rather than at the end of PEG chains.

[00233] In addition to these forms of PEG, the polymer can also be prepared with weak or degradable linkages in the backbone. For example, PEG can be prepared with ester linkages in the polymer backbone that are subject to hydrolysis. As shown herein, this hydrolysis results in cleavage of the polymer into fragments of lower molecular weight: -PEG-CO_2-PEG- $+H_2O \rightarrow$ PEG-CO_2H+HO-PEG- It is understood by those skilled in the art that the term poly(ethylene glycol) or PEG represents or includes all the forms known in the art including but not limited to those disclosed herein.

[00234] Many other polymers are also suitable for use. In some embodiments, polymer backbones that are water-soluble, with from 2 to about 300 termini, are particularly suitable. Examples of suitable polymers include, but are not limited to, other poly(alkylene glycols), such as poly(propylene glycol) ("PPG"), copolymers thereof (including but not limited to copolymers of ethylene glycol and propylene glycol), terpolymers thereof, mixtures thereof, and the like. Although the molecular weight of each chain of the polymer backbone can vary, it is typically in the range of from about 800 Da to about 100,000 Da, often from about 6,000 Da to about 80,000 Da.

[00235] Those of ordinary skill in the art will recognize that the foregoing list for substantially water soluble backbones is by no means exhaustive and is merely illustrative, and that all polymeric materials having the qualities described herein are contemplated as being suitable for use.

[00236] In some embodiments the polymer derivatives are "multi-functional", meaning that the polymer backbone has at least two termini, and possibly as many as about 300 termini, functionalized or activated with a functional group. Multifunctional polymer derivatives

include, but are not limited to, linear polymers having two termini, each terminus being bonded to a functional group which may be the same or different.

4. Linkers

[00237] In certain embodiments, the antibodies can be linked to the payloads with one or more linkers capable of reacting with an antibody amino acid and with a payload group. The one or more linkers can be any linkers apparent to those of skill in the art.

[00238] **The term "linker" is used herein to refer to groups or bonds that normally are formed** as the result of a chemical reaction and typically are covalent linkages.

[00239] Useful linkers include those described herein. In certain embodiments, the linker is any divalent or multivalent linker known to those of skill in the art. Useful divalent linkers include alkylene, substituted alkylene, heteroalkylene, substituted heteroalkylene, arylene, substituted arylene, heteroarlyene, and substituted heteroarylene. In certain embodiments, the linker is C_{1-10} alkylene or C_{1-10} heteroalkylene. In some embodiments, the C_{1-10}heteoalkylene is PEG.

[00240] In certain embodiments, the linker is hydrolytically stable. Hydrolytically stable linkages means that the linkages are substantially stable in water and do not react with water at useful pH values, including but not limited to, under physiological conditions for an extended period of time, perhaps even indefinitely. In certain embodiments, the linker is hydrolytically unstable. Hydrolytically unstable or degradable linkages mean that the linkages are degradable in water or in aqueous solutions, including for example, blood. Enzymatically unstable or degradable linkages mean that the linkage can be degraded by one or more enzymes.

[00241] As understood in the art, PEG and related polymers may include degradable linkages in the polymer backbone or in the linker group between the polymer backbone and one or more of the terminal functional groups of the polymer molecule. For example, ester linkages formed by the reaction of PEG carboxylic acids or activated PEG carboxylic acids with alcohol groups on a biologically active agent generally hydrolyze under physiological conditions to release the agent.

[00242] Other hydrolytically degradable linkages include, but are not limited to, carbonate linkages; imine linkages resulted from reaction of an amine and an aldehyde; phosphate ester linkages formed by reacting an alcohol with a phosphate group; hydrazone linkages which are reaction product of a hydrazide and an aldehyde; acetal linkages that are the reaction product

of an aldehyde and an alcohol; orthoester linkages that are the reaction product of a formate and an alcohol; peptide linkages formed by an amine group, including but not limited to, at an end of a polymer such as PEG, and a carboxyl group of a peptide; and oligonucleotide linkages formed by a phosphoramidite group, including but not limited to, at the end of a polymer, and a 5' hydroxyl group of an oligonucleotide.

[00243] A number of different cleavable linkers are known to those of skill in the art. See U.S. Pat. Nos. 4,618,492; 4,542,225, and 4,625,014. The mechanisms for release of an agent from these linker groups include, for example, irradiation of a photolabile bond and acid-catalyzed hydrolysis. U.S. Pat. No. 4,671,958, for example, includes a description of immunoconjugates comprising linkers which are cleaved at the target site in vivo by the proteolytic enzymes of the patient's complement system. The length of the linker may be predetermined or selected depending upon a desired spatial relationship between the polypeptide and the molecule linked to it. In view of the large number of methods that have been reported for attaching a variety of radiodiagnostic compounds, radiotherapeutic compounds, drugs, toxins, and other agents to polypeptides one skilled in the art will be able to determine a suitable method for attaching a given agent to a polypeptide.

[00244] The linker may have a wide range of molecular weight or molecular length. Larger or smaller molecular weight linkers may be used to provide a desired spatial relationship or conformation between the polypeptide and the linked entity. Linkers having longer or shorter molecular length may also be used to provide a desired space or flexibility between the polypeptide and the linked entity. Similarly, a linker having a particular shape or conformation may be utilized to impart a particular shape or conformation to the polypeptide or the linked entity, either before or after the polypeptide reaches its target. The functional groups present on each end of the linker may be selected to modulate the release of a polypeptide or a payload under desired conditions. This optimization of the spatial relationship between the polypeptide and the linked entity may provide new, modulated, or desired properties to the molecule.

[00245] In some embodiments, provided herein water-soluble bifunctional linkers that have a dumbbell structure that includes: a) an azide, an alkyne, a hydrazine, a hydrazide, a hydroxylamine, or a carbonyl-containing moiety on at least a first end of a polymer backbone; and b) at least a second functional group on a second end of the polymer backbone. The second functional group can be the same or different as the first functional group. The second functional group, in some embodiments, is not reactive with the first functional group. In some

embodiments, water-soluble compounds that comprise at least one arm of a branched molecular structure are provided. For example, the branched molecular structure can be a dendritic structure.

[00246] In some embodiments, the linker is derived from a linker precursor 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) or 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). In a specific embodiment, the linker is derived from the linker precursor N-succinimidyl 4-(maleimidomethyl)cyclohexanecarboxylate (SMCC).

[00247] In some embodiments, the linker is derived from a linker precursor selected from the group consisting of dipeptides, tripeptides, tetrapeptides, and pentapeptides. In such embodiments, the linker can be cleaved by a protease. Exemplary dipeptides include, but are not limited to, valine-citrulline (vc or val-cit), alanine-phenylalanine (af or ala-phe); phenylalanine-lysine (fk or phe-lys); phenylalanine-homolysine (phe-homolys); and N-methyl-valine-citrulline (Me-val-cit). Exemplary tripeptides include, but are not limited to, glycine-valine-citrulline (gly-val-cit), glycine-glycine-glycine (gly-gly-gly), and glycine-methoxyethoxyethyl)serine-valine (gly-val-citalanine OMESerValAla).

[00248] In some embodiments, a linker comprises a self-immolative spacer. In certain embodiments, the self-immolative spacer comprises p-aminobenzyl. In some embodiments, a p-aminobenzyl alcohol is attached to an amino acid unit via an amide bond, and a carbamate, methylcarbamate, or carbonate is made between the benzyl alcohol and the payload (Hamann *et al.* (2005) Expert Opin. Ther. Patents (2005) 15:1087-1103). In some embodiments, the linker comprises p-aminobenzyloxycarbonyl (PAB). Other examples of self-immolative spacers include, but are not limited to, aromatic compounds that are electronically similar to the PAB group, such as 2-aminoimidazol-5-methanol derivatives (U.S. Pat. No. 7,375,078; Hay *et al.* (1999) Bioorg. Med. Chem. Lett. 9:2237) and ortho- or para-aminobenzylacetals. In some embodiments, spacers can be used that undergo cyclization upon amide bond hydrolysis,

such as substituted and unsubstituted 4-aminobutyric acid amides (Rodrigues *et al.* (1995) Chemistry Biology 2:223), appropriately substituted bicyclo[2.2.1] and bicyclo[2.2.2] ring systems (Storm *et al.* (1972) J. Amer. Chem. Soc. 94:5815) and 2-aminophenylpropionic acid amides (Amsberry, *et al.* (1990) J. Org. Chem. 55:5867). Linkage of a drug to the α-carbon of a glycine residue is another example of a self-immolative spacer that may be useful in conjugates (Kingsbury *et al.* (1984) J. Med. Chem. 27:1447).

[00249] In certain embodiments, linker precursors can be combined to form larger linkers. For instance, in certain embodiments, linkers comprise the dipeptide valine-citrulline and p-aminobenzyloxycarbonyl. These are also referenced as citValCit--PAB linkers.

[00250] In certain embodiments, the payloads can be linked to the linkers, referred to herein as a linker-payload, with one or more linker groups capable of reacting with an antibody amino acid group. The one or more linkers can be any linkers apparent to those of skill in the art or those set forth herein.

[00251] Additional linkers are disclosed herein, such as, for example, the linker precursors (A) – (L) described below.

5. Antibody Specificity

[00252] The conjugates comprise antibodies that selectively bind human folate receptor alpha. In some aspects, the antibody selectively binds to the extracellular domain of human folate receptor alpha (human FOLR1).

[00253] In some embodiments, the antibody binds to a homolog of human FOLR1. In some aspects, the antibody binds to a homolog of human FOLR1 from a species selected from monkeys, mice, dogs, cats, rats, cows, horses, goats and sheep. In some aspects, the homolog is a cynomolgus monkey homolog. In some aspects, the homolog is a mouse or murine analog.

[00254] In some embodiments, the antibodies comprise at least one CDR sequence defined by a consensus sequence provided in this disclosure. In some embodiments, the antibodies comprise an illustrative CDR, V_H, or V_L sequence provided in this disclosure, or a variant thereof. In some aspects, the variant is a variant with a conservative amino acid substitution.

[00255] In some embodiments, the antibody has one or more CDRs having particular lengths, in terms of the number of amino acid residues. In some embodiments, the Chothia CDR-H1 of the antibody is 6, 7, or 8 residues in length. In some embodiments, the Kabat CDR-H1 of the antibody is 4, 5, or 6 residues in length. In some embodiments, the Chothia CDR-H2

of the antibody is 5, 6, or 7 residues in length. In some embodiments, the Kabat CDR-H2 of the antibody is 16, 17, or 18 residues in length. In some embodiments, the Kabat/Chothia CDR-H3 of the antibody is 13, 14, 15, 16, or 17 residues in length.

[00256] In some aspects, the Kabat/Chothia CDR-L1 of the antibody is 11, 12, 13, 14, 15, 16, 17, or 18 residues in length. In some aspects, the Kabat/Chothia CDR-L2 of the antibody is 6, 7, or 8 residues in length. In some aspects, the Kabat/Chothia CDR-L3 of the antibody is 8, 9, or 10 residues in length.

[00257] In some embodiments, the antibody comprises a light chain. In some aspects, the light chain is a kappa light chain. In some aspects, the light chain is a lambda light chain.

[00258] In some embodiments, the antibody comprises a heavy chain. In some aspects, the heavy chain is an IgA. In some aspects, the heavy chain is an IgD. In some aspects, the heavy chain is an IgE. In some aspects, the heavy chain is an IgG. In some aspects, the heavy chain is an IgM. In some aspects, the heavy chain is an IgG1. In some aspects, the heavy chain is an IgG2. In some aspects, the heavy chain is an IgG3. In some aspects, the heavy chain is an IgG4. In some aspects, the heavy chain is an IgA1. In some aspects, the heavy chain is an IgA2.

[00259] In some embodiments, the antibody is an antibody fragment. In some aspects, the antibody fragment is an Fv fragment. In some aspects, the antibody fragment is a Fab fragment. In some aspects, the antibody fragment is a $F(ab')_2$ fragment. In some aspects, the antibody fragment is a Fab' fragment. In some aspects, the antibody fragment is an scFv (sFv) fragment. In some aspects, the antibody fragment is an scFv-Fc fragment.

[00260] In some embodiments, the antibody is a monoclonal antibody. In some embodiments, the antibody is a polyclonal antibody.

[00261] In some embodiments, the antibody is a chimeric antibody. In some embodiments, the antibody is a humanized antibody. In some embodiments, the antibody is a human antibody.

[00262] In some embodiments, the antibody is an affinity matured antibody. In some aspects, the antibody is an affinity matured antibody derived from an illustrative sequence provided in this disclosure.

[00263] The antibodies provided herein may be useful for the treatment of a variety of diseases and conditions including cancers. In some embodiments, the antibodies provided herein may be useful for the treatment of cancers of solid tumors. For example, the antibodies provided herein can be useful for the treatment of colorectal cancer.

5.1 CDR-H3 Sequences

[00264] In some embodiments, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of a CDR-H3 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the CDR-H3 sequence is a CDR-H3 sequence of a V_H sequence provided in SEQ ID NOs.: 308-366.

[00265] In some embodiments, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs.: 240-298. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 240. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 241. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 242. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 243. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 244. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 245. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 246. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 247. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 248. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 249. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 250. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 251. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 252. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 253. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 254. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 255. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 256. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 257. In

some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 258. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 259. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 260. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 261. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 262. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 263. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 264. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 265. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 266. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 267. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 268. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 269. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 270. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 271. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 272. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 273. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 274. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 275. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 276. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 277. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 278. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 279. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of

SEQ ID NO: 280. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 281. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 282. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 283. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 284. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 285. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 286. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 287. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 288. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 289. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 290. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 291. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 292. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 293. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 294. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 295. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 296. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 297. In some aspects, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 298.

[00266] In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-H3 sequence provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-H3 sequences provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-H3 sequences provided in this

disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.2 V_H Sequences Comprising Illustrative CDRs

[00267] In some embodiments, the antibody comprises a V_H sequence comprising one or more CDR-H sequences comprising, consisting of, or consisting essentially of one or more illustrative CDR-H sequences provided in this disclosure, and variants thereof. In some embodiments, the CDR-H sequences comprise, consist of, or consist essentially of one or more CDR-H sequences provided in a V_H sequence selected from SEQ ID NOs: 308-366.

5.2.1. V_H Sequences Comprising Illustrative Kabat CDRs

[00268] In some embodiments, the antibody comprises a V_H sequence comprising one or more Kabat CDR-H sequences comprising, consisting of, or consisting essentially of one or more illustrative Kabat CDR-H sequences provided in this disclosure, and variants thereof.

5.2.1.1. Kabat CDR-H3

[00269] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H3 sequence, wherein the CDR-H3 sequence comprises, consists of, or consists essentially of a Kabat CDR-H3 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the Kabat CDR-H3 sequence is a Kabat CDR-H3 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00270] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs.: 240-298. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 240. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 241. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 242. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 243. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 244. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting

essentially of SEQ ID NO: 245. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 246. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 247. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 248. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 249. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 250. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 251. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 252. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 253. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 254. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 255. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 256. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 257. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 258. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 259. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 260. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 261. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 262. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting

essentially of SEQ ID NO: 263. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 264. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 265. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 266. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 267. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 268. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 269. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 270. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 271. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 272. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 273. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 274. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 275. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 276. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 277. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 278. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 279. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 280. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting

essentially of SEQ ID NO: 281. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 282. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 283. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 284. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 285. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 286. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 287. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 288. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 289. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 290. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 291. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 292. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 293. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 294. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 295. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 296. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 297. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 298.

5.2.1.2. Kabat CDR-H2

[00271] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H2 sequence, wherein the CDR-H2 sequence comprises, consists of, or consists essentially of a Kabat CDR-H2 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the Kabat CDR-H2 sequence is a Kabat CDR-H2 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00272] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-239. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 181. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 182. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 183. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 184. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 185. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 186. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 187. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 188. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 189. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 190. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 191. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 192. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 193. In some aspects, the antibody comprises a V_H sequence comprising a Kabat

CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 194. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 195. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 196. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 197. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 198. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 199. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 200. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 201. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 202. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 203. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 204. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 205. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 206. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 207. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 208. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 209. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 210. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 211. In some aspects, the antibody comprises a V_H sequence comprising a Kabat

CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 212. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 213. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 214. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 215. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 216. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 217. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 218. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 219. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 220. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 221. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 222. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 223. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 224. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 225. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 226. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 227. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 228. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 229. In some aspects, the antibody comprises a V_H sequence comprising a Kabat

CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 230. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 231. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 232. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 233. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 234. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 235. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 236. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 237. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 238. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 239.

5.2.1.3. Kabat CDR-H1

[00273] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H1 sequence, wherein the CDR-H1 sequence comprises, consists of, or consists essentially of a Kabat CDR-H1 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the Kabat CDR-H1 sequence is a Kabat CDR-H1 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00274] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 63-121. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 63. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 64. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 65. In some aspects, the

antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 66. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 67. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 68. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 69. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 70. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 71. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 72. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 73. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 74. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 75. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 76. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 77. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 78. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 79. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 80. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 81. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 82. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 83. In some aspects, the

antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 84. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 85. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 86. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 87. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 88. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 89. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 90. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 91. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 92. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 93. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 94. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 95. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 96. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 97. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 98. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 99. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 100. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 101. In some aspects, the

antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 102. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 103. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 104. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 105. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 106. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 107. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 108. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 109. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 110. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 111. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 112. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 113. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 114. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 115. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 116. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 117. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 118. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 119. In some aspects, the

antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 120. In some aspects, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 121.

5.2.1.4. Kabat CDR-H3 + Kabat CDR-H2

[00275] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298, and a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-239. In some aspects, the Kabat CDR-H3 sequence and the Kabat CDR-H2 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Kabat CDR-H3 and Kabat CDR-H2 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.1.5. Kabat CDR-H3 + Kabat CDR-H1

[00276] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298, and a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 63-121. In some aspects, the Kabat CDR-H3 sequence and the Kabat CDR-H1 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Kabat CDR-H3 and Kabat CDR-H1 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.1.6. Kabat CDR-H1 + Kabat CDR-H2

[00277] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 63-121 and a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-239. In some aspects, the Kabat CDR-H1 sequence and the Kabat CDR-H2 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Kabat CDR-H1 and Kabat CDR-H2 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.1.7. Kabat CDR-H1 + Kabat CDR-H2 + Kabat CDR-H3

[00278] In some embodiments, the antibody comprises a V_H sequence comprising a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 63-121, a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-239, and a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298. In some aspects, the Kabat CDR-H1 sequence, Kabat CDR-H2 sequence, and Kabat CDR-H3 sequence are all from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Kabat CDR-H1, Kabat CDR-H2, and Kabat CDR-H3 are all from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.1.8. Variants of V_H Sequences Comprising Illustrative Kabat CDRs

[00279] In some embodiments, the V_H sequences provided herein comprise a variant of an illustrative Kabat CDR-H3, CDR-H2, and/or CDR-H1 sequence provided in this disclosure.

[00280] In some aspects, the Kabat CDR-H3 sequence comprises, consists of, or consists essentially of a variant of an illustrative Kabat CDR-H3 sequence provided in this disclosure. In some aspects, the Kabat CDR-H3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Kabat CDR-H3 sequences provided in this disclosure. In some aspects, the Kabat CDR-H3 sequence comprises, consists of, or consists essentially of any of the illustrative Kabat CDR-H3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00281] In some aspects, the Kabat CDR-H2 sequence comprises, consists of, or consists essentially of a variant of an illustrative Kabat CDR-H2 sequence provided in this disclosure. In some aspects, the Kabat CDR-H2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Kabat CDR-H2 sequences provided in this disclosure. In some aspects, the Kabat CDR-H2 sequence comprises, consists of, or consists essentially of any of the illustrative Kabat CDR-H2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00282] In some aspects, the Kabat CDR-H1 sequence comprises, consists of, or consists essentially of a variant of an illustrative Kabat CDR-H1 sequence provided in this disclosure. In some aspects, the Kabat CDR-H1 sequence comprises, consists of, or consists essentially of

a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Kabat CDR-H1 sequences provided in this disclosure. In some aspects, the Kabat CDR-H1 sequence comprises, consists of, or consists essentially of any of the illustrative Kabat CDR-H1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.2.2. V_H Sequences Comprising Illustrative Chothia CDRs

[00283] In some embodiments, the antibody comprises a V_H sequence comprising one or more Chothia CDR-H sequences comprising, consisting of, or consisting essentially of one or more illustrative Chothia CDR-H sequences provided in this disclosure, and variants thereof.

5.2.2.1. Chothia CDR-H3

[00284] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H3 sequence, wherein the CDR-H3 sequence comprises, consists of, or consists essentially of a Chothia CDR-H3 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the Chothia CDR-H3 sequence is a Chothia CDR-H3 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00285] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 240. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 241. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 242. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 243. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 244. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 245. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 246. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 247. In

some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 248. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 249. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 250. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 251. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 252. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 253. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 254. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 255. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 256. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 257. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 258. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 259. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 260. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 261. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 262. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 263. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 264. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 265. In

some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 266. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 267. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 268. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 269. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 270. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 271. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 272. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 273. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 274. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 275. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 276. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 277. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 278. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 279. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 280. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 281. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 282. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 283. In

some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 284. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 285. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 286. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 287. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 288. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 289. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 290. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 291. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 292. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 293. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 294. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 295. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 296. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 297. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 298.

5.2.2.2. Chothia CDR-H2

[00286] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H2 sequence, wherein the CDR-H2 sequence comprises, consists of, or consists essentially of a Chothia CDR-H2 sequence of an illustrative antibody or V_H sequence provided herein. In

some aspects, the Chothia CDR-H2 sequence is a Chothia CDR-H2 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00287] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 122-180. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 122. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 123. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 124. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 125. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 126. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 127. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 128. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 129. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 130. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 131. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 132. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 133. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 134. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 135. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 136. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising,

consisting of, or consisting essentially of SEQ ID NO: 137. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 138. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 139. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 140. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 141. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 142. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 143. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 144. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 145. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 146. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 147. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 148. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 149. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 150. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 151. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 152. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 153. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 154. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising,

consisting of, or consisting essentially of SEQ ID NO: 155. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 156. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 157. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 158. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 159. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 160. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 161. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 162. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 163. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 164. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 165. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 166. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 167. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 168. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 169. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 170. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 171. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 172. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising,

consisting of, or consisting essentially of SEQ ID NO: 173. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 174. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 175. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 176. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 177. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 178. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 179. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 180.

5.2.2.3. Chothia CDR-H1

[00288] In some embodiments, the antibody comprises a V_H sequence comprising a CDR-H1 sequence, wherein the CDR-H1 sequence comprises, consists of, or consists essentially of a Chothia CDR-H1 sequence of an illustrative antibody or V_H sequence provided herein. In some aspects, the Chothia CDR-H1 sequence is a Chothia CDR-H1 sequence of a V_H sequence provided in SEQ ID NOs: 308-366.

[00289] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 4-62. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 4. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 5. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 6. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 7. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 8. In some aspects, the antibody comprises a V_H

sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 9. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 10. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 11. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 12. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 13. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 14. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 15. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 16. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 17. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 18. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 19. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 20. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 21. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 22. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 23. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 24. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 25. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 26. In some aspects, the antibody comprises a V_H

sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 27. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 28. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 29. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 30. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 31. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 32. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 33. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 34. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 35. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 36. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 37. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 38. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 39. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 40. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 41. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 42. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 43. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 44. In some aspects, the antibody comprises a V_H

sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 45. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 46. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 47. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 48. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 49. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 50. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 51. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 52. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 53. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 54. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 55. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 56. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 57. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 58. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 59. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 60. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 61. In some aspects, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 62.

5.2.2.4. Chothia CDR-H3 + Chothia CDR-H2

[00290] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298, and a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 122-180. In some aspects, the Chothia CDR-H3 sequence and the Chothia CDR-H2 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Chothia CDR-H3 and Chothia CDR-H2 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.2.5. Chothia CDR-H3 + Chothia CDR-H1

[00291] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298, and a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 4-62. In some aspects, the Chothia CDR-H3 sequence and the Chothia CDR-H1 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Chothia CDR-H3 and Chothia CDR-H1 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.2.6. Chothia CDR-H1 + Chothia CDR-H2

[00292] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 4-62 and a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 122-180. In some aspects, the Chothia CDR-H1 sequence and the Chothia CDR-H2 sequence are both from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Chothia CDR-H1 and Chothia CDR-H2 are both from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.2.7. Chothia CDR-H1 + Chothia CDR-H2 + Chothia CDR-H3

[00293] In some embodiments, the antibody comprises a V_H sequence comprising a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 4-62, a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 122-180, and a Chothia CDR-H3

sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 240-298. In some aspects, the Chothia CDR-H1 sequence, Chothia CDR-H2 sequence, and Chothia CDR-H3 sequence are all from a single illustrative V_H sequence provided in this disclosure. For example, in some aspects, the Chothia CDR-H1, Chothia CDR-H2, and Chothia CDR-H3 are all from a single illustrative V_H sequence selected from SEQ ID NOs: 308-366.

5.2.2.8. Variants of V_H Sequences Comprising Illustrative Chothia CDRs

[00294] In some embodiments, the V_H sequences provided herein comprise a variant of an illustrative Chothia CDR-H3, CDR-H2, and/or CDR-H1 sequence provided in this disclosure.

[00295] In some aspects, the Chothia CDR-H3 sequence comprises, consists of, or consists essentially of a variant of an illustrative Chothia CDR-H3 sequence provided in this disclosure. In some aspects, the Chothia CDR-H3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Chothia CDR-H3 sequences provided in this disclosure. In some aspects, the Chothia CDR-H3 sequence comprises, consists of, or consists essentially of any of the illustrative Chothia CDR-H3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00296] In some aspects, the Chothia CDR-H2 sequence comprises, consists of, or consists essentially of a variant of an illustrative Chothia CDR-H2 sequence provided in this disclosure. In some aspects, the Chothia CDR-H2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Chothia CDR-H2 sequences provided in this disclosure. In some aspects, the Chothia CDR-H2 sequence comprises, consists of, or consists essentially of any of the illustrative Chothia CDR-H2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00297] In some aspects, the Chothia CDR-H1 sequence comprises, consists of, or consists essentially of a variant of an illustrative Chothia CDR-H1 sequence provided in this disclosure. In some aspects, the Chothia CDR-H1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Chothia CDR-H1 sequences provided in this disclosure. In some aspects, the

Chothia CDR-H1 sequence comprises, consists of, or consists essentially of any of the illustrative Chothia CDR-H1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.3. V_H Sequences

[00298] In some embodiments, the antibody comprises, consists of, or consists essentially of a V_H sequence provided in SEQ ID NOs: 308-366.

[00299] In some embodiments, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 308-366. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 308. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 309. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 310. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 311. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 312. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 313. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 314. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 315. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 316. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 317. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 318. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 319. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 320. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 321. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 322. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 323. In some aspects, the antibody comprises a V_H sequence

comprising, consisting of, or consisting essentially of SEQ ID NO: 324. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 325. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 326. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 327. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 328. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 329. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 330. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 331. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 332. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 333. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 334. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 335. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 336. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 337. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 338. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 339. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 340. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 341. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 342. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 343. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 344. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 345. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 346. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 347. In some

aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 348. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 349. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 350. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 351. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 352. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 353. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 354. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 355. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 356. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 357. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 358. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 359. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 360. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 361. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 362. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 363. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 364. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 365. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 366.

5.3.1. Variants of V_H Sequences

[00300] In some embodiments, the V_H sequences provided herein comprise, consist of, or consist essentially of a variant of an illustrative V_H sequence provided in this disclosure.

[00301] In some aspects, the V_H sequence comprises, consists of, or consists essentially of a variant of an illustrative V_H sequence provided in this disclosure. In some aspects, the V_H

sequence comprises, consists of, or consists essentially of a sequence having at least 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 99.5% identity with any of the illustrative V_H sequences provided in this disclosure.

[00302] In some embodiments, the V_H sequence comprises, consists of, or consists essentially of any of the illustrative V_H sequences provided in this disclosure having 20 or fewer, 19 or fewer, 18 or fewer, 17 or fewer, 16 or fewer, 15 or fewer, 14 or fewer, 13 or fewer, 12 or fewer, 11 or fewer, 10 or fewer, 9 or fewer, 8 or fewer, 7 or fewer, 6 or fewer, 5 or fewer, 4 or fewer, 3 or fewer, 2 or fewer, or 1 or fewer amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.4. CDR-L3 Sequences

[00303] In some embodiments, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of a CDR-L3 sequence of an illustrative antibody or V_L sequence provided herein. In some aspects, the CDR-L3 sequence is a CDR-L3 sequence of a V_L sequence provided in SEQ ID NOs.: 367-369.

[00304] In some embodiments, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 305-307. In some aspects, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 305. In some aspects, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 306. In some aspects, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 307.

[00305] In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L3 sequence provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L3 sequences provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.5. V_L Sequences Comprising Illustrative CDRs

[00306] In some embodiments, the antibody comprises a V_L sequence comprising one or more CDR-L sequences comprising, consisting of, or consisting essentially of one or more illustrative CDR-L sequences provided in this disclosure, and variants thereof.

5.5.1. CDR-L3

[00307] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L3 sequence, wherein the CDR-L3 sequence comprises, consists of, or consists essentially of a CDR-L3 sequence of an illustrative antibody or V_L sequence provided herein. In some aspects, the CDR-L3 sequence is a CDR-L3 sequence of a V_L sequence provided in SEQ ID NOs.: 367-369.

[00308] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 305-307. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 305. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 306. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 307.

5.5.2. CDR-L2

[00309] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L2 sequence, wherein the CDR-L2 sequence comprises, consists of, or consists essentially of a CDR-L2 sequence of an illustrative antibody or V_L sequence provided herein. In some aspects, the CDR-L2 sequence is a CDR-L2 sequence of a V_L sequence provided in SEQ ID NOs.: 367-369.

[00310] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 302-304. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 302. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 303. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 304.

5.5.3. CDR-L1

[00311] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L1 sequence, wherein the CDR-L1 sequence comprises, consists of, or consists essentially of a CDR-L1 sequence of an illustrative antibody or V_L sequence provided herein. In some aspects, the CDR-L1 sequence is a CDR-L1 sequence of a V_L sequence provided in SEQ ID NOs.: 367-369.

[00312] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 299-301. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 299. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 300. In some aspects, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 301.

5.5.4. CDR-L3 + CDR-L2

[00313] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 305-307 and a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 302-304. In some aspects, the CDR-L3 sequence and the CDR-L2 sequence are both from a single illustrative V_L sequence provided in this disclosure. For example, in some aspects, the CDR-L3 and CDR-L2 are both from a single illustrative V_L sequence selected from SEQ ID NOs.: 367-369.

5.5.5. CDR-L3 + CDR-L1

[00314] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 305-307 and a CDR-L1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 299-301. In some aspects, the CDR-L3 sequence and the CDR-L1 sequence are both from a single illustrative V_L sequence provided in this disclosure. For example, in some aspects, the CDR-L3 and CDR-L1 are both from a single illustrative V_L sequence selected from SEQ ID NOs.: 367-369.

5.5.6. CDR-L1 + CDR-L2

[00315] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 299-301 and a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 302-304. In some aspects, the CDR-L1 sequence and the CDR-L2 sequence are both from a single illustrative V_L sequence provided in this disclosure. For example, in some aspects, the CDR-L1 and CDR-L2 are both from a single illustrative V_L sequence selected from SEQ ID NOs.: 367-369.

5.5.7. CDR-L1 + CDR-L2 + CDR-L3

[00316] In some embodiments, the antibody comprises a V_L sequence comprising a CDR-L1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 299-301, a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 302-304, and a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 305-307. In some aspects, the CDR-L1 sequence, CDR-L2 sequence, and CDR-L3 sequence are all from a single illustrative V_L sequence provided in this disclosure. For example, in some aspects, the CDR-L1, CDR-L2, and CDR-L3 are all from a single illustrative V_L sequence selected from SEQ ID NOs.: 367-369.

5.5.8. Variants of VL Sequences Comprising Illustrative CDR-Ls

[00317] In some embodiments, the V_L sequences provided herein comprise a variant of an illustrative CDR-L3, CDR-L2, and/or CDR-L1 sequence provided in this disclosure.

[00318] In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L3 sequence provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L3 sequences provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00319] In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L2 sequence provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a sequence

having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L2 sequences provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00320] In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L1 sequence provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L1 sequences provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.6. V_L Sequences

[00321] In some embodiments, the antibody comprises, consists of, or consists essentially of a V_L sequence provided in SEQ ID NOs.: 367-369.

[00322] In some embodiments, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs.: 367-369. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 367. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 368. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 369.

5.6.1. Variants of V_L Sequences

[00323] In some embodiments, the V_L sequences provided herein comprise, consist of, or consist essentially of a variant of an illustrative V_L sequence provided in this disclosure.

[00324] In some aspects, the V_L sequence comprises, consists of, or consists essentially of a variant of an illustrative V_L sequence provided in this disclosure. In some aspects, the V_L sequence comprises, consists of, or consists essentially of a sequence having at least 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 99.5% identity with any of the illustrative V_L sequences provided in this disclosure.

[00325] In some embodiments, the V_L sequence comprises, consists of, or consists essentially of any of the illustrative V_L sequences provided in this disclosure having 20 or fewer, 19 or fewer, 18 or fewer, 17 or fewer, 16 or fewer, 15 or fewer, 14 or fewer, 13 or fewer, 12 or fewer, 11 or fewer, 10 or fewer, 9 or fewer, 8 or fewer, 7 or fewer, 6 or fewer, 5 or fewer, 4 or fewer, 3 or fewer, 2 or fewer, or 1 or fewer amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.7. Pairs

5.7.1. CDR-H3 – CDR-L3 Pairs

[00326] In some embodiments, the antibody comprises a CDR-H3 sequence and a CDR-L3 sequence. In some aspects, the CDR-H3 sequence is part of a V_H and the CDR-L3 sequence is part of a V_L.

[00327] In some aspects, the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 240-298, and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00328] In some aspects, the CDR-H3 - CDR-L3 pairs are selected from SEQ ID NO: 305 and SEQ ID NO: 240; SEQ ID NO: 305 and SEQ ID NO: 241; SEQ ID NO: 305 and SEQ ID NO: 242; SEQ ID NO: 305 and SEQ ID NO: 243; SEQ ID NO: 305 and SEQ ID NO: 244; SEQ ID NO: 305 and SEQ ID NO: 245; SEQ ID NO: 305 and SEQ ID NO: 246; SEQ ID NO: 305 and SEQ ID NO: 247; SEQ ID NO: 305 and SEQ ID NO: 248; SEQ ID NO: 305 and SEQ ID NO: 249; SEQ ID NO: 305 and SEQ ID NO: 250; SEQ ID NO: 305 and SEQ ID NO: 251; SEQ ID NO: 305 and SEQ ID NO: 252; SEQ ID NO: 305 and SEQ ID NO: 253; SEQ ID NO: 305 and SEQ ID NO: 254; SEQ ID NO: 305 and SEQ ID NO: 255; SEQ ID NO: 305 and SEQ ID NO: 256; SEQ ID NO: 305 and SEQ ID NO: 257; SEQ ID NO: 305 and SEQ ID NO: 258; SEQ ID NO: 305 and SEQ ID NO: 259; SEQ ID NO: 305 and SEQ ID NO: 260; SEQ ID NO: 305 and SEQ ID NO: 261; SEQ ID NO: 305 and SEQ ID NO: 262; SEQ ID NO: 305 and SEQ ID NO: 263; SEQ ID NO: 305 and SEQ ID NO: 264; SEQ ID NO: 305 and SEQ ID NO: 265; SEQ ID NO: 305 and SEQ ID NO: 266; SEQ ID NO: 305 and SEQ ID NO: 267; SEQ ID NO: 305 and SEQ ID NO: 268; SEQ ID NO: 305 and SEQ ID NO: 269; SEQ ID NO: 305 and SEQ ID NO: 270; SEQ ID NO: 305 and SEQ ID NO: 271; SEQ ID NO: 305 and SEQ ID NO: 272; SEQ ID NO: 305 and SEQ ID NO: 273; SEQ ID NO: 305 and SEQ ID NO: 274; SEQ ID NO: 305 and SEQ ID NO: 275; SEQ ID NO: 305 and SEQ ID NO: 276; SEQ ID NO: 305 and SEQ

ID NO: 277; SEQ ID NO: 305 and SEQ ID NO: 278; SEQ ID NO: 305 and SEQ ID NO: 279; SEQ ID NO: 305 and SEQ ID NO: 280; SEQ ID NO: 305 and SEQ ID NO: 281; SEQ ID NO: 305 and SEQ ID NO: 282; SEQ ID NO: 305 and SEQ ID NO: 283; SEQ ID NO: 305 and SEQ ID NO: 284; SEQ ID NO: 305 and SEQ ID NO: 285; SEQ ID NO: 305 and SEQ ID NO: 286; SEQ ID NO: 305 and SEQ ID NO: 287; SEQ ID NO: 305 and SEQ ID NO: 288; SEQ ID NO: 305 and SEQ ID NO: 289; SEQ ID NO: 305 and SEQ ID NO: 290; SEQ ID NO: 305 and SEQ ID NO: 291; SEQ ID NO: 305 and SEQ ID NO: 292; SEQ ID NO: 305 and SEQ ID NO: 293; SEQ ID NO: 305 and SEQ ID NO: 294; SEQ ID NO: 305 and SEQ ID NO: 295; SEQ ID NO: 305 and SEQ ID NO: 296; SEQ ID NO: 305 and SEQ ID NO: 297; and SEQ ID NO: 305 and SEQ ID NO: 298.

[00329] In some aspects, the CDR-H3 - CDR-L3 pairs are selected from SEQ ID NO: 306 and SEQ ID NO: 240; SEQ ID NO: 306 and SEQ ID NO: 241; SEQ ID NO: 306 and SEQ ID NO: 242; SEQ ID NO: 306 and SEQ ID NO: 243; SEQ ID NO: 306 and SEQ ID NO: 244; SEQ ID NO: 306 and SEQ ID NO: 245; SEQ ID NO: 306 and SEQ ID NO: 246; SEQ ID NO: 306 and SEQ ID NO: 247; SEQ ID NO: 306 and SEQ ID NO: 248; SEQ ID NO: 306 and SEQ ID NO: 249; SEQ ID NO: 306 and SEQ ID NO: 250; SEQ ID NO: 306 and SEQ ID NO: 251; SEQ ID NO: 306 and SEQ ID NO: 252; SEQ ID NO: 306 and SEQ ID NO: 253; SEQ ID NO: 306 and SEQ ID NO: 254; SEQ ID NO: 306 and SEQ ID NO: 255; SEQ ID NO: 306 and SEQ ID NO: 256; SEQ ID NO: 306 and SEQ ID NO: 257; SEQ ID NO: 306 and SEQ ID NO: 258; SEQ ID NO: 306 and SEQ ID NO: 259; SEQ ID NO: 306 and SEQ ID NO: 260; SEQ ID NO: 306 and SEQ ID NO: 261; SEQ ID NO: 306 and SEQ ID NO: 262; SEQ ID NO: 306 and SEQ ID NO: 263; SEQ ID NO: 306 and SEQ ID NO: 264; SEQ ID NO: 306 and SEQ ID NO: 265; SEQ ID NO: 306 and SEQ ID NO: 266; SEQ ID NO: 306 and SEQ ID NO: 267; SEQ ID NO: 306 and SEQ ID NO: 268; SEQ ID NO: 306 and SEQ ID NO: 269; SEQ ID NO: 306 and SEQ ID NO: 270; SEQ ID NO: 306 and SEQ ID NO: 271; SEQ ID NO: 306 and SEQ ID NO: 272; SEQ ID NO: 306 and SEQ ID NO: 273; SEQ ID NO: 306 and SEQ ID NO: 274; SEQ ID NO: 306 and SEQ ID NO: 275; SEQ ID NO: 306 and SEQ ID NO: 276; SEQ ID NO: 306 and SEQ ID NO: 277; SEQ ID NO: 306 and SEQ ID NO: 278; SEQ ID NO: 306 and SEQ ID NO: 279; SEQ ID NO: 306 and SEQ ID NO: 280; SEQ ID NO: 306 and SEQ ID NO: 281; SEQ ID NO: 306 and SEQ ID NO: 282; SEQ ID NO: 306 and SEQ ID NO: 283; SEQ ID NO: 306 and SEQ ID NO: 284; SEQ ID NO: 306 and SEQ ID NO: 285; SEQ ID NO: 306 and SEQ ID NO: 286; SEQ ID NO: 306 and SEQ ID NO: 287; SEQ ID NO: 306 and SEQ ID NO: 288; SEQ ID NO: 306 and SEQ ID NO: 289; SEQ ID NO: 306 and SEQ ID NO: 290; SEQ ID NO: 306 and SEQ

ID NO: 291; SEQ ID NO: 306 and SEQ ID NO: 292; SEQ ID NO: 306 and SEQ ID NO: 293; SEQ ID NO: 306 and SEQ ID NO: 294; SEQ ID NO: 306 and SEQ ID NO: 295; SEQ ID NO: 306 and SEQ ID NO: 296; SEQ ID NO: 306 and SEQ ID NO: 297; and SEQ ID NO: 306 and SEQ ID NO: 298.

[00330] In some aspects, the CDR-H3 - CDR-L3 pairs are selected from SEQ ID NO: 307 and SEQ ID NO: 240; SEQ ID NO: 307 and SEQ ID NO: 241; SEQ ID NO: 307 and SEQ ID NO: 242; SEQ ID NO: 307 and SEQ ID NO: 243; SEQ ID NO: 307 and SEQ ID NO: 244; SEQ ID NO: 307 and SEQ ID NO: 245; SEQ ID NO: 307 and SEQ ID NO: 246; SEQ ID NO: 307 and SEQ ID NO: 247; SEQ ID NO: 307 and SEQ ID NO: 248; SEQ ID NO: 307 and SEQ ID NO: 249; SEQ ID NO: 307 and SEQ ID NO: 250; SEQ ID NO: 307 and SEQ ID NO: 251; SEQ ID NO: 307 and SEQ ID NO: 252; SEQ ID NO: 307 and SEQ ID NO: 253; SEQ ID NO: 307 and SEQ ID NO: 254; SEQ ID NO: 307 and SEQ ID NO: 255; SEQ ID NO: 307 and SEQ ID NO: 256; SEQ ID NO: 307 and SEQ ID NO: 257; SEQ ID NO: 307 and SEQ ID NO: 258; SEQ ID NO: 307 and SEQ ID NO: 259; SEQ ID NO: 307 and SEQ ID NO: 260; SEQ ID NO: 307 and SEQ ID NO: 261; SEQ ID NO: 307 and SEQ ID NO: 262; SEQ ID NO: 307 and SEQ ID NO: 263; SEQ ID NO: 307 and SEQ ID NO: 264; SEQ ID NO: 307 and SEQ ID NO: 265; SEQ ID NO: 307 and SEQ ID NO: 266; SEQ ID NO: 307 and SEQ ID NO: 267; SEQ ID NO: 307 and SEQ ID NO: 268; SEQ ID NO: 307 and SEQ ID NO: 269; SEQ ID NO: 307 and SEQ ID NO: 270; SEQ ID NO: 307 and SEQ ID NO: 271; SEQ ID NO: 307 and SEQ ID NO: 272; SEQ ID NO: 307 and SEQ ID NO: 273; SEQ ID NO: 307 and SEQ ID NO: 274; SEQ ID NO: 307 and SEQ ID NO: 275; SEQ ID NO: 307 and SEQ ID NO: 276; SEQ ID NO: 307 and SEQ ID NO: 277; SEQ ID NO: 307 and SEQ ID NO: 278; SEQ ID NO: 307 and SEQ ID NO: 279; SEQ ID NO: 307 and SEQ ID NO: 280; SEQ ID NO: 307 and SEQ ID NO: 281; SEQ ID NO: 307 and SEQ ID NO: 282; SEQ ID NO: 307 and SEQ ID NO: 283; SEQ ID NO: 307 and SEQ ID NO: 284; SEQ ID NO: 307 and SEQ ID NO: 285; SEQ ID NO: 307 and SEQ ID NO: 286; SEQ ID NO: 307 and SEQ ID NO: 287; SEQ ID NO: 307 and SEQ ID NO: 288; SEQ ID NO: 307 and SEQ ID NO: 289; SEQ ID NO: 307 and SEQ ID NO: 290; SEQ ID NO: 307 and SEQ ID NO: 291; SEQ ID NO: 307 and SEQ ID NO: 292; SEQ ID NO: 307 and SEQ ID NO: 293; SEQ ID NO: 307 and SEQ ID NO: 294; SEQ ID NO: 307 and SEQ ID NO: 295; SEQ ID NO: 307 and SEQ ID NO: 296; SEQ ID NO: 307 and SEQ ID NO: 297; and SEQ ID NO: 307 and SEQ ID NO: 298.

5.7.1.1. Variants of CDR-H3 – CDR-L3 Pairs

[00331] In some embodiments, the CDR-H3 – CDR-L3 pairs provided herein comprise a variant of an illustrative CDR-H3 and/or CDR-L1 sequence provided in this disclosure.

[00332] In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-H3 sequence provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-H3 sequences provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-H3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00333] In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L3 sequence provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L3 sequences provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.7.2. CDR-H1 – CDR-L1 Pairs

[00334] In some embodiments, the antibody comprises a CDR-H1 sequence and a CDR-L1 sequence. In some aspects, the CDR-H1 sequence is part of a V_H and the CDR-L1 sequence is part of a V_L.

[00335] In some aspects, the CDR-H1 sequence is a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 4-62, and the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301.

[00336] In some aspects, the CDR-H1 sequence is a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 63-121, and the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301.

5.7.2.1. Variants of CDR-H1 – CDR-L1 Pairs

[00337] In some embodiments, the CDR-H1 – CDR-L1 pairs provided herein comprise a variant of an illustrative CDR-H1 and/or CDR-L1 sequence provided in this disclosure.

[00338] In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-H1 sequence provided in this disclosure. In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-H1 sequences provided in this disclosure. In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-H1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00339] In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L1 sequence provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L1 sequences provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.7.3. CDR-H2 – CDR-L2 Pairs

[00340] In some embodiments, the antibody comprises a CDR-H2 sequence and a CDR-L2 sequence. In some aspects, the CDR-H2 sequence is part of a V_H and the CDR-L2 sequence is part of a V_L.

[00341] In some aspects, the CDR-H2 sequence is a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 122-180, and the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304.

[00342] In some aspects, the CDR-H1 sequence is a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 181-239, and the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304.

5.7.3.1. Variants of CDR-H2 – CDR-L2 Pairs

[00343] In some embodiments, the CDR-H2 – CDR-L2 pairs provided herein comprise a variant of an illustrative CDR-H2 and/or CDR-L2 sequence provided in this disclosure.

[00344] In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-H2 sequence provided in this disclosure. In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-H2 sequences provided in this disclosure. In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-H2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00345] In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L2 sequence provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L2 sequences provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.7.4. $V_H – V_L$ Pairs

[00346] In some embodiments, the antibody comprises a V_H sequence and a V_L sequence.

[00347] In some aspects, the V_H sequence is a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 308-366, and the V_L sequence is a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 367-369.

[00348] In some aspects, the V_H - V_L pairs are selected from SEQ ID NO: 367 and SEQ ID NO: 308; SEQ ID NO: 367 and SEQ ID NO: 309; SEQ ID NO: 367 and SEQ ID NO: 310; SEQ ID NO: 367 and SEQ ID NO: 311; SEQ ID NO: 367 and SEQ ID NO: 312; SEQ ID NO: 367 and SEQ ID NO: 313; SEQ ID NO: 367 and SEQ ID NO: 314; SEQ ID NO: 367 and SEQ ID NO: 315; SEQ ID NO: 367 and SEQ ID NO: 316; SEQ ID NO: 367 and SEQ ID NO: 317; SEQ ID NO: 367 and SEQ ID NO: 318; SEQ ID NO: 367 and SEQ ID NO: 319; SEQ ID NO: 367 and SEQ ID NO: 320; SEQ ID NO: 367 and SEQ ID NO: 321; SEQ ID NO: 367 and SEQ ID NO: 322; SEQ ID NO: 367 and SEQ ID NO: 323; SEQ ID NO: 367 and SEQ ID NO: 324;

SEQ ID NO: 367 and SEQ ID NO: 325; SEQ ID NO: 367 and SEQ ID NO: 326; SEQ ID NO: 367 and SEQ ID NO: 327; SEQ ID NO: 367 and SEQ ID NO: 328; SEQ ID NO: 367 and SEQ ID NO: 329; SEQ ID NO: 367 and SEQ ID NO: 330; SEQ ID NO: 367 and SEQ ID NO: 331; SEQ ID NO: 367 and SEQ ID NO: 332; SEQ ID NO: 367 and SEQ ID NO: 333; SEQ ID NO: 367 and SEQ ID NO: 334; SEQ ID NO: 367 and SEQ ID NO: 335; SEQ ID NO: 367 and SEQ ID NO: 336; SEQ ID NO: 367 and SEQ ID NO: 337; SEQ ID NO: 367 and SEQ ID NO: 338; SEQ ID NO: 367 and SEQ ID NO: 339; SEQ ID NO: 367 and SEQ ID NO: 340; SEQ ID NO: 367 and SEQ ID NO: 341; SEQ ID NO: 367 and SEQ ID NO: 342; SEQ ID NO: 367 and SEQ ID NO: 343; SEQ ID NO: 367 and SEQ ID NO: 344; SEQ ID NO: 367 and SEQ ID NO: 345; SEQ ID NO: 367 and SEQ ID NO: 346; SEQ ID NO: 367 and SEQ ID NO: 347; SEQ ID NO: 367 and SEQ ID NO: 348; SEQ ID NO: 367 and SEQ ID NO: 349; SEQ ID NO: 367 and SEQ ID NO: 350; SEQ ID NO: 367 and SEQ ID NO: 351; SEQ ID NO: 367 and SEQ ID NO: 352; SEQ ID NO: 367 and SEQ ID NO: 353; SEQ ID NO: 367 and SEQ ID NO: 354; SEQ ID NO: 367 and SEQ ID NO: 355; SEQ ID NO: 367 and SEQ ID NO: 356; SEQ ID NO: 367 and SEQ ID NO: 357; SEQ ID NO: 367 and SEQ ID NO: 358; SEQ ID NO: 367 and SEQ ID NO: 359; SEQ ID NO: 367 and SEQ ID NO: 360; SEQ ID NO: 367 and SEQ ID NO: 361; SEQ ID NO: 367 and SEQ ID NO: 362; SEQ ID NO: 367 and SEQ ID NO: 363; SEQ ID NO: 367 and SEQ ID NO: 364; SEQ ID NO: 367 and SEQ ID NO: 365; and SEQ ID NO: 367 and SEQ ID NO: 366.

[00349] In some aspects, the V_H - V_L pairs are selected from SEQ ID NO: 368 and SEQ ID NO: 308; SEQ ID NO: 368 and SEQ ID NO: 309; SEQ ID NO: 368 and SEQ ID NO: 310; SEQ ID NO: 368 and SEQ ID NO: 311; SEQ ID NO: 368 and SEQ ID NO: 312; SEQ ID NO: 368 and SEQ ID NO: 313; SEQ ID NO: 368 and SEQ ID NO: 314; SEQ ID NO: 368 and SEQ ID NO: 315; SEQ ID NO: 368 and SEQ ID NO: 316; SEQ ID NO: 368 and SEQ ID NO: 317; SEQ ID NO: 368 and SEQ ID NO: 318; SEQ ID NO: 368 and SEQ ID NO: 319; SEQ ID NO: 368 and SEQ ID NO: 320; SEQ ID NO: 368 and SEQ ID NO: 321; SEQ ID NO: 368 and SEQ ID NO: 322; SEQ ID NO: 368 and SEQ ID NO: 323; SEQ ID NO: 368 and SEQ ID NO: 324; SEQ ID NO: 368 and SEQ ID NO: 325; SEQ ID NO: 368 and SEQ ID NO: 326; SEQ ID NO: 368 and SEQ ID NO: 327; SEQ ID NO: 368 and SEQ ID NO: 328; SEQ ID NO: 368 and SEQ ID NO: 329; SEQ ID NO: 368 and SEQ ID NO: 330; SEQ ID NO: 368 and SEQ ID NO: 331; SEQ ID NO: 368 and SEQ ID NO: 332; SEQ ID NO: 368 and SEQ ID NO: 333; SEQ ID NO: 368 and SEQ ID NO: 334; SEQ ID NO: 368 and SEQ ID NO: 335; SEQ ID NO: 368 and SEQ ID NO: 336; SEQ ID NO: 368 and SEQ ID NO: 337; SEQ ID NO: 368 and SEQ ID NO: 338;

SEQ ID NO: 368 and SEQ ID NO: 339; SEQ ID NO: 368 and SEQ ID NO: 340; SEQ ID NO: 368 and SEQ ID NO: 341; SEQ ID NO: 368 and SEQ ID NO: 342; SEQ ID NO: 368 and SEQ ID NO: 343; SEQ ID NO: 368 and SEQ ID NO: 344; SEQ ID NO: 368 and SEQ ID NO: 345; SEQ ID NO: 368 and SEQ ID NO: 346; SEQ ID NO: 368 and SEQ ID NO: 347; SEQ ID NO: 368 and SEQ ID NO: 348; SEQ ID NO: 368 and SEQ ID NO: 349; SEQ ID NO: 368 and SEQ ID NO: 350; SEQ ID NO: 368 and SEQ ID NO: 351; SEQ ID NO: 368 and SEQ ID NO: 352; SEQ ID NO: 368 and SEQ ID NO: 353; SEQ ID NO: 368 and SEQ ID NO: 354; SEQ ID NO: 368 and SEQ ID NO: 355; SEQ ID NO: 368 and SEQ ID NO: 356; SEQ ID NO: 368 and SEQ ID NO: 357; SEQ ID NO: 368 and SEQ ID NO: 358; SEQ ID NO: 368 and SEQ ID NO: 359; SEQ ID NO: 368 and SEQ ID NO: 360; SEQ ID NO: 368 and SEQ ID NO: 361; SEQ ID NO: 368 and SEQ ID NO: 362; SEQ ID NO: 368 and SEQ ID NO: 363; SEQ ID NO: 368 and SEQ ID NO: 364; SEQ ID NO: 368 and SEQ ID NO: 365; and SEQ ID NO: 368 and SEQ ID NO: 366.

[00350] In some aspects, the V_H - V_L pairs are selected from SEQ ID NO: 369 and SEQ ID NO: 308; SEQ ID NO: 369 and SEQ ID NO: 309; SEQ ID NO: 369 and SEQ ID NO: 310; SEQ ID NO: 369 and SEQ ID NO: 311; SEQ ID NO: 369 and SEQ ID NO: 312; SEQ ID NO: 369 and SEQ ID NO: 313; SEQ ID NO: 369 and SEQ ID NO: 314; SEQ ID NO: 369 and SEQ ID NO: 315; SEQ ID NO: 369 and SEQ ID NO: 316; SEQ ID NO: 369 and SEQ ID NO: 317; SEQ ID NO: 369 and SEQ ID NO: 318; SEQ ID NO: 369 and SEQ ID NO: 319; SEQ ID NO: 369 and SEQ ID NO: 320; SEQ ID NO: 369 and SEQ ID NO: 321; SEQ ID NO: 369 and SEQ ID NO: 322; SEQ ID NO: 369 and SEQ ID NO: 323; SEQ ID NO: 369 and SEQ ID NO: 324; SEQ ID NO: 369 and SEQ ID NO: 325; SEQ ID NO: 369 and SEQ ID NO: 326; SEQ ID NO: 369 and SEQ ID NO: 327; SEQ ID NO: 369 and SEQ ID NO: 328; SEQ ID NO: 369 and SEQ ID NO: 329; SEQ ID NO: 369 and SEQ ID NO: 330; SEQ ID NO: 369 and SEQ ID NO: 331; SEQ ID NO: 369 and SEQ ID NO: 332; SEQ ID NO: 369 and SEQ ID NO: 333; SEQ ID NO: 369 and SEQ ID NO: 334; SEQ ID NO: 369 and SEQ ID NO: 335; SEQ ID NO: 369 and SEQ ID NO: 336; SEQ ID NO: 369 and SEQ ID NO: 337; SEQ ID NO: 369 and SEQ ID NO: 338; SEQ ID NO: 369 and SEQ ID NO: 339; SEQ ID NO: 369 and SEQ ID NO: 340; SEQ ID NO: 369 and SEQ ID NO: 341; SEQ ID NO: 369 and SEQ ID NO: 342; SEQ ID NO: 369 and SEQ ID NO: 343; SEQ ID NO: 369 and SEQ ID NO: 344; SEQ ID NO: 369 and SEQ ID NO: 345; SEQ ID NO: 369 and SEQ ID NO: 346; SEQ ID NO: 369 and SEQ ID NO: 347; SEQ ID NO: 369 and SEQ ID NO: 348; SEQ ID NO: 369 and SEQ ID NO: 349; SEQ ID NO: 369 and SEQ ID NO: 350; SEQ ID NO: 369 and SEQ ID NO: 351; SEQ ID NO: 369 and SEQ ID NO: 352;

SEQ ID NO: 369 and SEQ ID NO: 353; SEQ ID NO: 369 and SEQ ID NO: 354; SEQ ID NO: 369 and SEQ ID NO: 355; SEQ ID NO: 369 and SEQ ID NO: 356; SEQ ID NO: 369 and SEQ ID NO: 357; SEQ ID NO: 369 and SEQ ID NO: 358; SEQ ID NO: 369 and SEQ ID NO: 359; SEQ ID NO: 369 and SEQ ID NO: 360; SEQ ID NO: 369 and SEQ ID NO: 361; SEQ ID NO: 369 and SEQ ID NO: 362; SEQ ID NO: 369 and SEQ ID NO: 363; SEQ ID NO: 369 and SEQ ID NO: 364; SEQ ID NO: 369 and SEQ ID NO: 365; and SEQ ID NO: 369 and SEQ ID NO: 366.

5.7.4.1. Variants of V_H – V_L Pairs

[00351] In some embodiments, the V_H – V_L pairs provided herein comprise a variant of an illustrative V_H and/or V_L sequence provided in this disclosure.

[00352] In some aspects, the V_H sequence comprises, consists of, or consists essentially of a variant of an illustrative V_H sequence provided in this disclosure. In some aspects, the V_H sequence comprises, consists of, or consists essentially of a sequence having at least 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 99.1% identity with any of the illustrative V_H sequences provided in this disclosure.

[00353] In some embodiments, the V_H sequence comprises, consists of, or consists essentially of any of the illustrative V_H sequences provided in this disclosure having 20 or fewer, 19 or fewer, 18 or fewer, 17 or fewer, 16 or fewer, 15 or fewer, 14 or fewer, 13 or fewer, 12 or fewer, 11 or fewer, 10 or fewer, 9 or fewer, 8 or fewer, 7 or fewer, 6 or fewer, 5 or fewer, 4 or fewer, 3 or fewer, 2 or fewer, or 1 or fewer amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00354] In some aspects, the V_L sequence comprises, consists of, or consists essentially of a variant of an illustrative V_L sequence provided in this disclosure. In some aspects, the V_L sequence comprises, consists of, or consists essentially of a sequence having at least 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 99.5% identity with any of the illustrative V_L sequences provided in this disclosure.

[00355] In some embodiments, the V_L sequence comprises, consists of, or consists essentially of any of the illustrative V_L sequences provided in this disclosure having 20 or fewer, 19 or fewer, 18 or fewer, 17 or fewer, 16 or fewer, 15 or fewer, 14 or fewer, 13 or fewer, 12 or fewer, 11 or fewer, 10 or fewer, 9 or fewer, 8 or fewer, 7 or fewer, 6 or fewer, 5 or fewer, 4 or fewer, 3 or fewer, 2 or fewer, or 1 or fewer amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.8. Antibodies Comprising All Six CDRs

[00356] In some embodiments, the antibody comprises a CDR-H1 sequence, a CDR-H2 sequence, a CDR-H3 sequence, a CDR-L1 sequence, and a CDR-L3 sequence. In some aspects, the CDR sequences are part of a V_H (for CDR-H) or V_L (for CDR-L).

[00357] In some aspects, the CDR-H1 sequence is a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 4-62; the CDR-H2 sequence is a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 122-180; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 240-298; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00358] In some aspects, the CDR-H1 sequence is a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 19; the CDR-H2 sequence is a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 137; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 255; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00359] In some aspects, the CDR-H1 sequence is a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 58; the CDR-H2 sequence is a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 176; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 294; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00360] In some aspects, the CDR-H1 sequence is a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 63-121; the CDR-H2 sequence is a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 181-239; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 240-298; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00361] In some aspects, the CDR-H1 sequence is a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 78; the CDR-H2 sequence is a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 196; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 255; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

[00362] In some aspects, the CDR-H1 sequence is a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 117; the CDR-H2 sequence is a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 235; the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 294; the CDR-L1 sequence is a CDR-L1 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 299-301; the CDR-L2 sequence is a CDR-L2 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 302-304; and the CDR-L3 sequence is a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 305-307.

5.8.1. Variants of Antibodies Comprising All Six CDRs

[00363] In some embodiments, the CDR-H1, CDR-H2, CDR-H3, CDR-L1, CDR-L2, and CDR-L3 provided herein comprise a variant of an illustrative CDR-H1, CDR-H2, CDR-H3, CDR-L1, CDR-L2, and/or CDR-L3 sequence provided in this disclosure.

[00364] In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of a variant of an illustrative Chothia or Kabat CDR-H1 sequence provided in this disclosure. In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Chothia or Kabat CDR-H1 sequences provided in this disclosure. In some aspects, the CDR-H1 sequence comprises, consists of, or consists essentially of any of the illustrative Chothia or Kabat CDR-H1 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00365] In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of a variant of an illustrative Chothia or Kabat CDR-H2 sequence provided in this disclosure. In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative Chothia or Kabat CDR-H2 sequences provided in this disclosure. In some aspects, the CDR-H2 sequence comprises, consists of, or consists essentially of any of the illustrative Chothia or Kabat CDR-H2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00366] In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-H3 sequence provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-H3 sequences provided in this disclosure. In some aspects, the CDR-H3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-H3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00367] In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L1 sequence provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L1 sequences provided in this disclosure. In some aspects, the CDR-L1 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L1 sequences provided in this

disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00368] In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L2 sequence provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L2 sequences provided in this disclosure. In some aspects, the CDR-L2 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L2 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

[00369] In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a variant of an illustrative CDR-L3 sequence provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of a sequence having at least 70%, 75%, 80%, 85%, 90%, or 95% identity with any of the illustrative CDR-L3 sequences provided in this disclosure. In some aspects, the CDR-L3 sequence comprises, consists of, or consists essentially of any of the illustrative CDR-L3 sequences provided in this disclosure, with 1, 2, or 3 amino acid substitutions. In some aspects, the amino acid substitutions are conservative amino acid substitutions.

5.9. Consensus Sequences

[00370] In some embodiments, provided herein are anti-FOLR1 antibodies comprising one or more sequences defined by consensus sequences. Each consensus sequence is based, at least in part, on one or more alignments of two or more useful anti-FOLR1 CDR sequences provided in this disclosure. Based on such alignments, a person of skill in the art would recognize that different amino acid residues may useful in certain positions of the CDRs. Accordingly, each consensus sequence encompasses two or more useful anti-FOLR1 CDR sequences.

[00371] In some embodiments, the antibodies comprise one to six of the consensus CDR sequences provided herein. In some embodiments, the antibodies comprise two to six of the consensus CDR sequences provided herein. In some embodiments, the antibodies comprise three to six of the consensus CDR sequences provided herein. In some embodiments, the antibodies comprise four to six of the consensus CDR sequences provided herein. In some embodiments, the antibodies comprise five to six of the consensus CDR sequences provided herein. In some embodiments, the antibodies comprise six of the consensus CDR sequences

provided herein. In some embodiments, the antibodies comprise a V_L comprising the CDR-L consensus sequence(s). In some embodiments, the antibodies comprise a V_H comprising the CDR-H consensus sequence(s). In some embodiments, the antibodies comprise a V_H comprising the CDR-H consensus sequence(s) and a V_L comprising the CDR-L consensus sequence(s).

5.9.1. CDR-H3 Consensus Sequences

[00372] In some embodiments, the antibody comprises a CDR-H3 sequence defined by the consensus sequence G-α_2-α_3-α_4-W-α_6-α_7-G-α_9-α_{10}-Y-α_{12}-α_{13}-α_{14}-Y, where α_2 is G, S, A, F, H, R, T, or Y; α_3 is W, L, or Y; α_4 is S, A, F, Y, H, or D; α_6 is R, P, Q, or K; α_7 is S, A, or H; α_9 is Y, H, or M; α_{10} is G, S, D, or W; α_{12} is Y or F; α_{13} is L, I, Q, or M; and α_{14} is D or E.

[00373] In some embodiments, the antibody comprises a CDR-H3 sequence defined by the consensus sequence G-α_2-α_3-α_4-W-α_6-α_7-G-α_9-α_{10}-Y-α_{12}-α_{13}-α_{14}-Y, where α_2 is G or S; α_3 is W; α_4 is S or H; α_6 is R or P; α_7 is S; α_9 is Y or M; α_{10} is G, S, or D; α_{12} is Y; α_{13} is L; and α_{14} is D.

5.9.2. Chothia CDR-H1 Consensus Sequences

[00374] In some embodiments, the antibody comprises a Chothia CDR-H1 sequence defined by the consensus sequence γ_1-γ_2-γ_3-γ_4-γ_5-γ_6-γ_7, where γ_1 is G or S; γ_2 is F or S; γ_3 is N; γ_4 is I or T; γ_5 is S, R, G, T, N, or D; γ_6 is N, K, T, R, H, Y, L, M, Q, or V; and γ_7 is Y, H, S, N, K, F, or Q.

[00375] In some embodiments, the antibody comprises a Chothia CDR-H1 sequence defined by the consensus sequence γ_1-γ_2-γ_3-γ_4-γ_5-γ_6-γ_7, where γ_1 is G; γ_2 is F; γ_3 is N; γ_4 is I or T; γ_5 is S, R, or T; γ_6 is N or T; and γ_7 is Y, K, or Q.

5.9.3. Chothia CDR-H2 Consensus Sequences

[00376] In some embodiments, the antibody comprises a Chothia CDR-H2 sequence defined by the consensus sequence ε_1-ε_2-ε_3-ε_4-ε_5-ε_6, where ε_1 is Y, T, F, S, or A; ε_2 is P; ε_3 is N, I, V, R, Y, F, G, L, Q, or S; ε_4 is D or P; ε_5 is G or D; and ε_6 is Y, I, T, N, F, S, or M.

[00377] In some embodiments, the antibody comprises a Chothia CDR-H2 sequence defined by the consensus sequence ε_1-ε_2-ε_3-ε_4-ε_5-ε_6, where ε_1 is Y or F; ε_2 is P; ε_3 is N, I, or R; ε_4 is D; ε_5 is G; and ε_6 is Y or I.

5.9.4. Kabat CDR-H1 Consensus Sequences

[00378] In some embodiments, the antibody comprises a Kabat CDR-H1 sequence defined by the consensus sequence ζ_1-ζ_2-ζ_3-ζ_4-ζ_5, where ζ_1 is N, K, T, R, H, Y, L, M, Q, or V; ζ_2 is Y, H, S, N, K, F, or Q; ζ_3 is S or Y; ζ_4 is I; and ζ_5 is H.

[00379] In some embodiments, the antibody comprises a Kabat CDR-H1 sequence defined by the consensus sequence ζ_1-ζ_2-ζ_3-ζ_4-ζ_5, where ζ_1 is N or T; ζ_2 is Y, K, or Q; ζ_3 is S; ζ_4 is I; and ζ_5 is H.

5.9.5. Kabat CDR-H2 Consensus Sequences

[00380] In some embodiments, the antibody comprises a Kabat CDR-H2 sequence defined by the consensus sequence θ_1-θ_2-θ_3-θ_4-θ_5-θ_6-θ_7-θ_8-θ_9-D-Y-A-D-θ_{14}-θ_{15}-θ_{16}-G, where θ_1 is G, E, D, W, S, or V; θ_2 is I or V; θ_3 is Y, T, F, S, or A; θ_4 is P; θ_5 is N, I, V, R, Y, F, G, L, Q, or S; θ_6 is D or P; θ_7 is G or D; θ_8 is Y, I, T, N, F, S, or M; θ_9 is T or N; θ_{14} is S, R, or N; θ_{15} is V or M; and θ_{16} is K or E.

[00381] In some embodiments, the antibody comprises a Kabat CDR-H2 sequence defined by the consensus sequence θ_1-θ_2-θ_3-θ_4-θ_5-θ_6-θ_7-θ_8-θ_9-D-Y-A-D-θ_{14}-θ_{15}-θ_{16}-G, where θ_1 is G, E, or D; θ_2 is I; θ_3 is Y or F; θ_4 is P; θ_5 is N, I, or R; θ_6 is D; θ_7 is G; θ_8 is Y or I; θ_9 is T; θ_{14} is S; θ_{15} is V; and θ_{16} is K.

6. Germline

[00382] In some embodiments, the antibody that specifically binds folate receptor alpha is an antibody comprising a variable region that is encoded by a particular germline gene, or a variant thereof. The illustrative antibodies provided herein comprise variable regions that are encoded by the heavy chain variable region germline genes VH1-18, VH3-33, VH2-5, VH2-70, and VH4-30-4. or variants thereof; and the light chain variable region germline genes Vκ1-5, Vκ3-11, Vκ2-20, Vκ1-33, and Vκ1-16, or variants thereof.

[00383] One of skill in the art would recognize that the CDR sequences provided herein may also be useful when combined with variable regions encoded by other variable region germline genes, or variants thereof. In particular, the CDR sequences provided herein may be useful when combined with variable regions encoded by variable region germline genes, or variants thereof, that are structurally similar to the variable region germline genes recited above. For example, in some embodiments, a CDR-H sequence provided herein may be combined with a variable region encoded by a variable region germline gene selected from the V_H 1, V_H 2, V_H

3, or V_H 4 families, or a variant thereof. In some embodiments, a CDR-L sequence provided herein may be combined with a variable region encoded by a variable region germline gene selected from the **Vκ1, Vκ2, or Vκ3, or a variant thereof.**

7. Affinity

[00384] In some embodiments, the affinity of the antibody for folate receptor alpha as indicated by K_D, is less than about 10^{-5} M, less than about 10^{-6} M, less than about 10^{-7} M, less than about 10^{-8} M, less than about 10^{-9} M, less than about 10^{-10} M, less than about 10^{-11} M, or less than about 10^{-12} M. In some embodiments, the affinity of the antibody is between about 10^{-7} M and 10^{-11} M. In some embodiments, the affinity of the antibody is between about 10^{-7} M and 10^{-10} M. In some embodiments, the affinity of the antibody is between about 10^{-7} M and 10^{-9} M. In some embodiments, the affinity of the antibody is between about 10^{-7} M and 10^{-8} M. In some embodiments, the affinity of the antibody is between about 10^{-8} M and 10^{-11} M. In some embodiments, the affinity of the antibody is between about 10^{-8} M and 10^{-10} M. In some embodiments, the affinity of the antibody is between about 10^{-9} M and 10^{-11} M. In some embodiments, the affinity of the antibody is between about 10^{-9} M and 10^{-10} M.

[00385] In some embodiments, the affinity of the antibody for human folate receptor alpha, as determined by surface plasmon resonance at 25°C, and as indicated by K_D, is from about 0.36×10^{-9} M to about 2.21×10^{-9} M. In some embodiments, the affinity of the antibody for human folate receptor alpha, as determined by surface plasmon resonance at 25°C, and as indicated by K_D, is from about 8.55×10^{-10} M to about 1.70×10^{-8} M. In some embodiments, the affinity of the antibody for human folate receptor alpha, as determined by surface plasmon resonance at 25°C, and as indicated by K_D, is from about 5.71×10^{-10} M to about 2.58×10^{-8} M. In some embodiments, the affinity of the antibody for human folate receptor alpha is about any of the K_D values reported for human folate receptor alpha in the examples below.

[00386] In some embodiments the antibody has a k_a of at least about 10^4 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of at least about 10^5 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of at least about 10^6 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of at least about 10^7 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of at least about 10^8 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of at least about 10^9 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of between about 10^4 $M^{-1}\times sec^{-1}$ and about 10^{10} $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of between about 10^5 $M^{-1}\times sec^{-1}$ and about 10^{10} $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of between about 10^6 $M^{-1}\times sec^{-1}$ and

about 10^{10} $M^{-1}\times sec^{-1}$. . In some embodiments the antibody has a k_a of between about 10^7 $M^{-1}\times sec^{-1}$ and about 10^{10} $M^{-1}\times sec^{-1}$.

[00387] In some embodiments the antibody has a k_a when associating with human folate receptor alpha, as determined by surface plasmon resonance at 25°C, of from about 4.44×10^5 $M^{-1}\times sec^{-1}$ to about 1.61×10^5 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a when associating with human folate receptor alpha, as determined by surface plasmon resonance at 25°C, of from about 2.90×10^5 $M^{-1}\times sec^{-1}$ to about 9.64×10^9 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a when associating with human folate receptor alpha of about any of the k_a values reported for human folate receptor alpha in the examples below.

[00388] In some embodiments the antibody has a k_d of about 10^{-5} sec^{-1} or less. In some embodiments the antibody has a k_d of about 10^{-4} sec^{-1} or less. In some embodiments the antibody has a k_d of about 10^{-3} sec^{-1} or less. In some embodiments the antibody has a k_d of between about 10^{-2} sec^{-1} and about 10^{-5} sec^{-1}. In some embodiments the antibody has a k_d of between about 10^{-2} sec^{-1} and about 10^{-4} sec^{-1}. In some embodiments the antibody has a k_d of between about 10^{-3} sec^{-1} and about 10^{-5} sec^{-1}.

[00389] In some embodiments the antibody has a k_d when dissociating from human folate receptor alpha, as determined by surface plasmon resonance at 25°C, of from about 8.66×10^{-4} sec^{-1} to about 1.08×10^{-2} sec^{-1}. In some embodiments the antibody has a k_d when dissociating from human folate receptor alpha, as determined by surface plasmon resonance at 25°C, of from about 2.28×10^{-4} sec^{-1} to about 4.82×10^{1} sec^{-1}. In some embodiments the antibody has a k_d when dissociating from human folate receptor alpha of about any of the k_d values reported for human folate receptor alpha in the examples below.

[00390] In some embodiments, the affinity of the antibody for cynomolgus folate receptor alpha, as determined by surface plasmon resonance at 25°C, and as indicated by K_D, is from about 0.19×10^{-9} M to about 2.84×10^{-9} M. In some embodiments, the affinity of the antibody for cynomolgus folate receptor alpha is about any of the K_D values reported for cynomolgus folate receptor alpha in the examples below.

[00391] In some embodiments, the affinity of the antibody for mouse folate receptor alpha, as determined by surface plasmon resonance at 25°C, and as indicated by K_D, is from about 0.5×10^{-9} M to about 9.07×10^{-8} M. In some embodiments, the affinity of the antibody for mouse folate receptor alpha is about any of the K_D values reported for mouse folate receptor alpha in the examples below.

[00392] In some aspects, the K_D, k_a, and k_d are determined at 25°C. In some embodiments, the K_D, k_a, and k_d are determined by surface plasmon resonance. In some embodiments, the K_D, k_a, and k_d are determined according to the methods described in the Examples provided herein.

8. Epitope Bins

[00393] In some embodiments, the antibody binds the same epitope as an antibody encompassing any of SEQ ID NOs: 308-366. In some embodiments, the antibody binds the same epitope as an antibody comprising any of the V_H-V_L pairs, above. In some embodiments, the antibody competes for epitope binding with an antibody encompassing any of SEQ ID NOs: 308-366. In some embodiments, the antibody competes for epitope binding with an antibody comprising any of the V_H-V_L pairs, above.

9. Glycosylation Variants

[00394] In certain embodiments, an antibody may be altered to increase, decrease or eliminate the extent to which it is glycosylated. Glycosylation of polypeptides is typically either "N-linked" or "O-linked."

[00395] "N-linked" glycosylation refers to the attachment of a carbohydrate moiety to the side chain of an asparagine residue. The tripeptide sequences asparagine-X-serine and asparagine-X-threonine, where X is any amino acid except proline, are the recognition sequences for enzymatic attachment of the carbohydrate moiety to the asparagine side chain. Thus, the presence of either of these tripeptide sequences in a polypeptide creates a potential glycosylation site.

[00396] "O-linked" glycosylation refers to the attachment of one of the sugars N-acetylgalactosamine, galactose, or xylose to a hydroxyamino acid, most commonly serine or threonine, although 5-hydroxyproline or 5-hydroxylysine may also be used.

[00397] Addition or deletion of N-linked glycosylation sites to the antibody may be accomplished by altering the amino acid sequence such that one or more of the above-described tripeptide sequences is created or removed. Addition or deletion of O-linked glycosylation sites may be accomplished by addition, deletion, or substitution of one or more serine or threonine residues in or to (as the case may be) the sequence of an antibody.

10. Fc Variants

[00398] In certain embodiments, amino acid modifications may be introduced into the Fc region of an antibody provided herein to generate an Fc region variant. In certain embodiments,

the Fc region variant possesses some, but not all, effector functions. Such antibodies may be useful, for example, in applications in which the half-life of the antibody *in vivo* is important, yet certain effector functions are unnecessary or deleterious. Examples of effector functions include complement-dependent cytotoxicity (CDC) and antibody-directed complement-mediated cytotoxicity (ADCC). Numerous substitutions or substitutions or deletions with altered effector function are known in the art.

[00399] In some embodiments, the Fc comprises one or more modifications in at least one of the C_H3 sequences. In some embodiments, the Fc comprises one or more modifications in at least one of the C_H2 sequences. For example, the Fc can include one or modifications selected from the group consisting of: V262E, V262D, V262K, V262R, V262S, V264S, V303R, and V305R. In some embodiments, an Fc is a single polypeptide. In some embodiments, an Fc is multiple peptides, *e.g.*, two polypeptides. Exemplary modifications in the Fc region are described, for example, in International Patent Application No. PCT/US2017/037545, filed June 14, 2017.

[00400] An alteration in in CDC and/or ADCC activity can be confirmed using *in vitro* and/or *in vivo* assays. For example, Fc receptor (FcR) binding assays can be conducted to measure FcγR binding. The primary cells for mediating ADCC, NK cells, express FcγRIII only, whereas monocytes express FcγRI, FcγRII and FcγRIII. FcR expression on hematopoietic cells is summarized in Ravetch and Kinet, *Ann. Rev. Immunol.*, 1991, 9:457-492, incorporated by reference in its entirety.

[00401] Non-limiting examples of *in vitro* assays to assess ADCC activity of a molecule of interest are provided in U.S. Patent Nos. 5,500,362 and 5,821,337; Hellstrom et al., *Proc. Natl. Acad. Sci. U.S.A.*, 1986, 83:7059-7063; Hellstrom et al., *Proc. Natl. Acad. Sci. U.S.A.*, 1985, 82:1499-1502; and Bruggemann et al., *J. Exp. Med.*, 1987, 166:1351-1361; each of which is incorporated by reference in its entirety. Useful effector cells for such assays include peripheral blood mononuclear cells (PBMC) and Natural Killer (NK) cells. Alternatively, or additionally, ADCC activity of the molecule of interest may be assessed *in vivo*, using an animal model such as that disclosed in Clynes et al. *Proc. Natl. Acad. Sci. U.S.A.*, 1998, 95:652-656, incorporated by reference in its entirety.

[00402] C1q binding assays may also be carried out to confirm that the antibody is unable to bind C1q and hence lacks CDC activity. Examples of C1q binding assays include those

described in WO 2006/029879 and WO 2005/100402, each of which is incorporated by reference in its entirety.

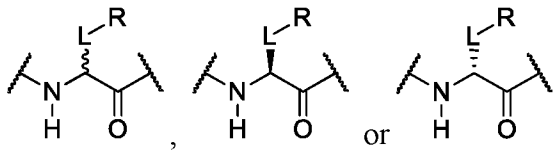
[00403] Complement activation assays include those described, for example, in Gazzano-Santoro et al., *J. Immunol. Methods*, 1996, 202:163-171; Cragg et al., *Blood*, 2003, 101:1045-1052; and Cragg and Glennie, *Blood*, 2004, 103:2738-2743; each of which is incorporated by reference in its entirety.

[00404] FcRn binding and *in vivo* clearance (half-life determination) can also be measured, for example, using the methods described in Petkova et al., *Intl. Immunol.*, 2006, 18:1759-1769, incorporated by reference in its entirety.

11. Modified Amino Acids

[00405] When the antibody conjugate comprises a modified amino acid, the modified amino acid can be any modified amino acid deemed suitable by the practitioner. In particular embodiments, the modified amino acid comprises a reactive group useful for forming a covalent bond to a linker precursor or to a payload precursor. In certain embodiments, the modified amino acid is a non-natural amino acid. In certain embodiments, the reactive group is selected from the group consisting of amino, carboxy, acetyl, hydrazino, hydrazido, semicarbazido, sulfanyl, azido and alkynyl. Modified amino acids are also described in, for example, WO 2013/185115 and WO 2015/006555, each of which is incorporated herein by reference in its entirety.

[00406] In certain embodiments, the amino acid residue is according to any of the following formulas:



Those of skill in the art will recognize that antibodies are generally comprised of L-amino acids However, with non-natural amino acids, the present methods and compositions provide the practitioner with the ability to use L-, D- or racemic non-natural amino acids at the site-specific positions. In certain embodiments, the non-natural amino acids described herein include D- versions of the natural amino acids and racemic versions of the natural amino acids.

[00407] In the above formulas, the wavy lines indicate bonds that connect to the remainder of the polypeptide chains of the antibodies. These non-natural amino acids can be incorporated

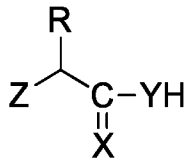
into polypeptide chains just as natural amino acids are incorporated into the same polypeptide chains. In certain embodiments, the non-natural amino acids are incorporated into the polypeptide chain via amide bonds as indicated in the formulas.

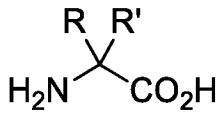
[00408] In the above formulas R designates any functional group without limitation, so long as the amino acid residue is not identical to a natural amino acid residue. In certain embodiments, R can be a hydrophobic group, a hydrophilic group, a polar group, an acidic group, a basic group, a chelating group, a reactive group, a therapeutic moiety or a labeling moiety. In certain embodiments, R is selected from the group consisting of $R^1NR^2R^3$, $R^1C(=O)R^2$, $R^1C(=O)OR^2$, R^1N_3, $R^1C(\equiv CH)$. In these embodiments, R^1 is selected from the group consisting of a bond, alkylene, heteroalkylene, arylene, heteroarylene. R^2 and R^3 are each independently selected from the group consisting of hydrogen, alkyl and heteroalkyl.

[00409] In some embodiments, the non-naturally encoded amino acids include side chain functional groups that react efficiently and selectively with functional groups not found in the 20 common amino acids (including but not limited to, azido, ketone, aldehyde and aminooxy groups) to form stable conjugates. For example, antigen-binding polypeptide that includes a non-naturally encoded amino acid containing an azido functional group can be reacted with a polymer (including but not limited to, poly(ethylene glycol) or, alternatively, a second polypeptide containing an alkyne moiety to form a stable conjugate resulting for the selective reaction of the azide and the alkyne functional groups to form a Huisgen [3+2] cycloaddition product.

[00410] Exemplary non-naturally encoded amino acids that may be suitable for use in the present invention and that are useful for reactions with water soluble polymers include, but are not limited to, those with carbonyl, aminooxy, hydrazine, hydrazide, semicarbazide, azide and alkyne reactive groups. In some embodiments, non-naturally encoded amino acids comprise a saccharide moiety. Examples of such amino acids include N-acetyl-L-glucosaminyl-L-serine, N-acetyl-L-galactosaminyl-L-serine, N-acetyl-L-glucosaminyl-L-threonine, N-acetyl-L-glucosaminyl-L-asparagine and O-mannosaminyl-L-serine. Examples of such amino acids also include examples where the naturally-occurring N– or O-linkage between the amino acid and the saccharide is replaced by a covalent linkage not commonly found in nature–including but not limited to, an alkene, an oxime, a thioether, an amide and the like. Examples of such amino acids also include saccharides that are not commonly found in naturally-occurring proteins such as 2-deoxy-glucose, 2-deoxygalactose and the like.

[00411] Many of the non-naturally encoded amino acids provided herein are commercially available, *e.g.*, from Sigma-Aldrich (St. Louis, Mo., USA), Novabiochem (a division of EMD Biosciences, Darmstadt, Germany), or Peptech (Burlington, Mass., USA). Those that are not commercially available are optionally synthesized as provided herein or using standard methods known to those of skill in the art. For organic synthesis techniques, see, *e.g.*, Organic Chemistry by Fessendon and Fessendon, (1982, Second Edition, Willard Grant Press, Boston Mass.); Advanced Organic Chemistry by March (Third Edition, 1985, Wiley and Sons, New York); and Advanced Organic Chemistry by Carey and Sundberg (Third Edition, Parts A and B, 1990, Plenum Press, New York). See, also, U.S. Patent Application Publications 2003/0082575 and 2003/0108885, which is incorporated by reference herein. In addition to unnatural amino acids that contain unnatural side chains, unnatural amino acids that may be suitable for use in the present invention also optionally comprise modified backbone structures, including but not limited to, as illustrated by the structures of Formula II and III:





wherein Z typically comprises OH, NH_2, SH, NH–R', or S–R'; X and Y, which can be the same or different, typically comprise S or O, and R and R', which are optionally the same or different, are typically selected from the same list of constituents for the R group described above for the unnatural amino acids having Formula I as well as hydrogen. For example, unnatural amino acids of the invention optionally comprise substitutions in the amino or carboxyl group as illustrated by Formulas II and III. Unnatural amino acids of this type include, but are not limited to, α-hydroxy acids, α-thioacids, α-aminothiocarboxylates, including but not limited to, with side chains corresponding to the common twenty natural amino acids or unnatural side chains. In addition, substitutions at the α-carbon optionally include, but are not limited to, L, D, or α-α-disubstituted amino acids such as D-glutamate, D-alanine, D-methyl-O-tyrosine, aminobutyric acid, and the like. Other structural alternatives include cyclic amino acids, such as proline analogues as well as 3, 4, 6, 7, 8, and 9 membered ring proline analogues, P and y amino acids such as substituted β-alanine and γ-amino butyric acid.

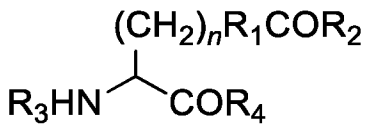
[00412] Many unnatural amino acids are based on natural amino acids, such as tyrosine, glutamine, phenylalanine, and the like, and are suitable for use in the present invention. Tyrosine analogs include, but are not limited to, para-substituted tyrosines, ortho-substituted tyrosines, and meta substituted tyrosines, where the substituted tyrosine comprises, including but not limited to, a keto group (including but not limited to, an acetyl group), a benzoyl group, an amino group, a hydrazine, an hydroxyamine, a thiol group, a carboxy group, an isopropyl group, a methyl group, a C_6-C_{20} straight chain or branched hydrocarbon, a saturated or unsaturated hydrocarbon, an O-methyl group, a polyether group, a nitro group, an alkynyl group or the like. In addition, multiply substituted aryl rings are also contemplated. Glutamine analogs that may be suitable for use in the present invention include, but are not limited to, α-hydroxy derivatives, γ-substituted derivatives, cyclic derivatives, and amide substituted glutamine derivatives. Example phenylalanine analogs that may be suitable for use in the present invention include, but are not limited to, para-substituted phenylalanines, ortho-substituted phenyalanines, and meta-substituted phenylalanines, where the substituent comprises, including but not limited to, a hydroxy group, a methoxy group, a methyl group, an allyl group, an aldehyde, an azido, an iodo, a bromo, a keto group (including but not limited to, an acetyl group), a benzoyl, an alkynyl group, or the like. Specific examples of unnatural amino acids that may be suitable for use in the present invention include, but are not limited to, a p-acetyl-L-phenylalanine, an O-methyl-L-tyrosine, an L-3-(2-naphthyl)alanine, a 3-methyl-phenylalanine, an O-4-allyl-L-tyrosine, a 4-propyl-L-tyrosine, a tri-O-acetyl-GlcNAcβ-serine, an L-Dopa, a fluorinated phenylalanine, an isopropyl-L-phenylalanine, a p-azido-L-phenylalanine, a p-azido-methyl-L-phenylalanine, a p-acyl-L-phenylalanine, a p-benzoyl-L-phenylalanine, an L-phosphoserine, a phosphonoserine, a phosphonotyrosine, a p-iodo-phenylalanine, a p-bromophenylalanine, a p-amino-L-phenylalanine, an isopropyl-L-phenylalanine, and a p-propargyloxy-phenylalanine, and the like. Examples of structures of a variety of unnatural amino acids that may be suitable for use in the present invention are provided in, for example, WO 2002/085923 entitled "In vivo incorporation of unnatural amino acids." See also Kiick et al., (2002) Incorporation of azides into recombinant proteins for chemoselective modification by the Staudinger ligation, PNAS 99:19-24, for additional methionine analogs.

[00413] Many of the unnatural amino acids suitable for use in the present invention are commercially available, *e.g.*, from Sigma (USA) or Aldrich (Milwaukee, Wis., USA). Those that are not commercially available are optionally synthesized as provided herein or as provided

in various publications or using standard methods known to those of skill in the art. For organic synthesis techniques, see, *e.g.*, Organic Chemistry by Fessendon and Fessendon, (1982, Second Edition, Willard Grant Press, Boston Mass.); Advanced Organic Chemistry by March (Third Edition, 1985, Wiley and Sons, New York); and Advanced Organic Chemistry by Carey and Sundberg (Third Edition, Parts A and B, 1990, Plenum Press, New York). Additional publications describing the synthesis of unnatural amino acids include, *e.g.*, WO 2002/085923 entitled "In vivo incorporation of Unnatural Amino Acids;" Matsoukas et al., (1995) J. Med. Chem., 38, 4660-4669; King, F. E. & Kidd, D. A. A. (1949) A New Synthesis of Glutamine and of γ-Dipeptides of Glutamic Acid from Phthylated Intermediates. J. Chem. Soc., 3315-3319; Friedman, O. M. & Chatterrji, R. (1959) Synthesis of Derivatives of Glutamine as Model Substrates for Anti-Tumor Agents. J. Am. Chem. Soc. 81, 3750-3752; Craig, J. C. et al. (1988) Absolute Configuration of the Enantiomers of 7-Chloro-4 [[4-(diethylamino)-1-methylbutyl]amino]quinoline (Chloroquine). J. Org. Chem. 53, 1167-1170; Azoulay, M., Vilmont, M. & Frappier, F. (1991) Glutamine analogues as Potential Antimalarials, Eur. J. Med. Chem. 26, 201-5; Koskinen, A. M. P. & Rapoport, H. (1989) Synthesis of 4-Substituted Prolines as Conformationally Constrained Amino Acid Analogues. J. Org. Chem. 54, 1859-1866; Christie, B. D. & Rapoport, H. (1985) Synthesis of Optically Pure Pipecolates from L-Asparagine. Application to the Total Synthesis of (+)-Apovincamine through Amino Acid Decarbonylation and Iminium Ion Cyclization. J. Org. Chem. 1989:1859-1866; Barton et al., (1987) Synthesis of Novel a-Amino-Acids and Derivatives Using Radical Chemistry: Synthesis of L- and D-a-Amino-Adipic Acids, L-a-aminopimelic Acid and Appropriate Unsaturated Derivatives. Tetrahedron Lett. 43:4297-4308; and, Subasinghe et al., (1992) Quisqualic acid analogues: synthesis of beta-heterocyclic 2-aminopropanoic acid derivatives and their activity at a novel quisqualate-sensitized site. J. Med. Chem. 35:4602-7. See also, patent applications entitled "Protein Arrays," filed Dec. 22, 2003, Ser. No. 10/744,899 and Ser. No. 60/435,821 filed on Dec. 22, 2002.

[00414] Amino acids with a carbonyl reactive group allow for a variety of reactions to link molecules (including but not limited to, PEG or other water soluble molecules) via nucleophilic addition or aldol condensation reactions among others.

[00415] Exemplary carbonyl-containing amino acids can be represented as follows:



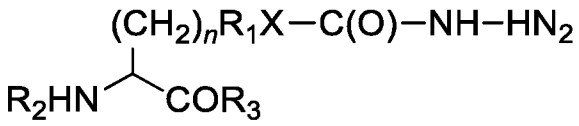
wherein n is 0-10; R_1 is an alkyl, aryl, substituted alkyl, or substituted aryl; R_2 is H, alkyl, aryl, substituted alkyl, and substituted aryl; and R_3 is H, an amino acid, a polypeptide, or an amino terminus modification group, and R_4 is H, an amino acid, a polypeptide, or a carboxy terminus modification group. In some embodiments, n is 1, R_1 is phenyl and R_2 is a simple alkyl (*i.e.*, methyl, ethyl, or propyl) and the ketone moiety is positioned in the para position relative to the alkyl side chain. In some embodiments, n is 1, R_1 is phenyl and R_2 is a simple alkyl (*i.e.*, methyl, ethyl, or propyl) and the ketone moiety is positioned in the meta position relative to the alkyl side chain.

[00416] In some examples, a non-naturally encoded amino acid bearing adjacent hydroxyl and amino groups can be incorporated into the polypeptide as a "masked" aldehyde functionality. For example, 5-hydroxylysine bears a hydroxyl group adjacent to the epsilon amine. Reaction conditions for generating the aldehyde typically involve addition of molar excess of sodium metaperiodate under mild conditions to avoid oxidation at other sites within the polypeptide. The pH of the oxidation reaction is typically about 7.0. A typical reaction involves the addition of about 1.5 molar excess of sodium meta periodate to a buffered solution of the polypeptide, followed by incubation for about 10 minutes in the dark. See, *e.g.* U.S. Pat. No. 6,423,685, which is incorporated by reference herein.

[00417] The carbonyl functionality can be reacted selectively with a hydrazine-, hydrazide-, hydroxylamine-, or semicarbazide-containing reagent under mild conditions in aqueous solution to form the corresponding hydrazone, oxime, or semicarbazone linkages, respectively, that are stable under physiological conditions. See, *e.g.*, Jencks, W. P., J. Am. Chem. Soc. 81, 475-481 (1959); Shao, J. and Tam, J. P., J. Am. Chem. Soc. 117:3893-3899 (1995). Moreover, the unique reactivity of the carbonyl group allows for selective modification in the presence of the other amino acid side chains. See, *e.g.*, Cornish, V. W., et al., J. Am. Chem. Soc. 118:8150-8151 (1996); Geoghegan, K. F. & Stroh, J. G., Bioconjug. Chem. 3:138-146 (1992); Mahal, L. K., et al., Science 276:1125-1128 (1997).

[00418] Non-naturally encoded amino acids containing a nucleophilic group, such as a hydrazine, hydrazide or semicarbazide, allow for reaction with a variety of electrophilic groups to form conjugates (including but not limited to, with PEG or other water soluble polymers).

[00419] Exemplary hydrazine, hydrazide or semicarbazide -containing amino acids can be represented as follows:



wherein n is 0-10; R_1 is an alkyl, aryl, substituted alkyl, or substituted aryl or not present; X, is O, N, or S or not present; R_2 is H, an amino acid, a polypeptide, or an amino terminus modification group, and R_3 is H, an amino acid, a polypeptide, or a carboxy terminus modification group.

[00420] In some embodiments, n is 4, R_1 is not present, and X is N. In some embodiments, n is 2, R_1 is not present, and X is not present. In some embodiments, n is 1, R_1 is phenyl, X is O, and the oxygen atom is positioned para to the aliphatic group on the aryl ring.

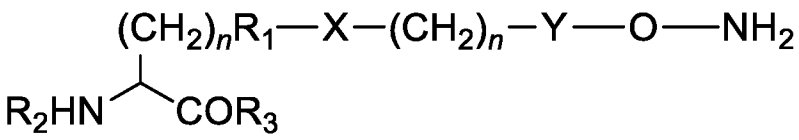
[00421] Hydrazide-, hydrazine-, and semicarbazide-containing amino acids are available from commercial sources. For instance, L-glutamate-γ-hydrazide is available from Sigma Chemical (St. Louis, Mo.). Other amino acids not available commercially can be prepared by one skilled in the art. See, *e.g.*, U.S. Pat. No. 6,281,211, which is incorporated by reference herein.

[00422] Polypeptides containing non-naturally encoded amino acids that bear hydrazide, hydrazine or semicarbazide functionalities can be reacted efficiently and selectively with a variety of molecules that contain aldehydes or other functional groups with similar chemical reactivity. See, *e.g.*, Shao, J. and Tam, J., J. Am. Chem. Soc. 117:3893-3899 (1995). The unique reactivity of hydrazide, hydrazine and semicarbazide functional groups makes them significantly more reactive toward aldehydes, ketones and other electrophilic groups as compared to the nucleophilic groups present on the 20 common amino acids (including but not limited to, the hydroxyl group of serine or threonine or the amino groups of lysine and the N-terminus).

[00423] Non-naturally encoded amino acids containing an aminooxy (also called a hydroxylamine) group allow for reaction with a variety of electrophilic groups to form conjugates (including but not limited to, with PEG or other water soluble polymers). Like hydrazines, hydrazides and semicarbazides, the enhanced nucleophilicity of the aminooxy group permits it to react efficiently and selectively with a variety of molecules that contain aldehydes or other functional groups with similar chemical reactivity. See, *e.g.*, Shao, J. and Tam, J., J. Am. Chem. Soc. 117:3893-3899 (1995); H. Hang and C. Bertozzi, Acc. Chem. Res. 34: 727-736 (2001). Whereas the result of reaction with a hydrazine group is the corresponding

hydrazone, however, an oxime results generally from the reaction of an aminooxy group with a carbonyl-containing group such as a ketone.

[00424] Exemplary amino acids containing aminooxy groups can be represented as follows:



wherein n is 0-10; R_1 is an alkyl, aryl, substituted alkyl, or substituted aryl or not present; X is O, N, S or not present; m is 0-10; Y=C(O) or not present; R_2 is H, an amino acid, a polypeptide, or an amino terminus modification group, and R_3 is H, an amino acid, a polypeptide, or a carboxy terminus modification group. In some embodiments, n is 1, R_1 is phenyl, X is O, m is 1, and Y is present. In some embodiments, n is 2, R_1 and X are not present, m is 0, and Y is not present.

[00425] Aminooxy-containing amino acids can be prepared from readily available amino acid precursors (homoserine, serine and threonine). See, *e.g.*, M. Carrasco and R. Brown, J. Org. Chem. 68: 8853-8858 (2003). Certain aminooxy-containing amino acids, such as L-2-amino-4-(aminooxy)butyric acid), have been isolated from natural sources (Rosenthal, G. et al., Life Sci. 60: 1635-1641 (1997). Other aminooxy-containing amino acids can be prepared by one skilled in the art.

[00426] The unique reactivity of azide and alkyne functional groups makes them extremely useful for the selective modification of polypeptides and other biological molecules. Organic azides, particularly aliphatic azides, and alkynes are generally stable toward common reactive chemical conditions. In particular, both the azide and the alkyne functional groups are inert toward the side chains (*i.e.*, R groups) of the 20 common amino acids found in naturally-occurring polypeptides. When brought into close proximity, however, the "spring-loaded" nature of the azide and alkyne groups is revealed and they react selectively and efficiently via Huisgen [3+2] cycloaddition reaction to generate the corresponding triazole. See, *e.g.*, Chin J., et al., Science 301:964-7 (2003); Wang, Q., et al., J. Am. Chem. Soc. 125, 3192-3193 (2003); Chin, J. W., et al., J. Am. Chem. Soc. 124:9026-9027 (2002).

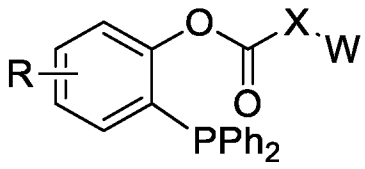
[00427] Because the Huisgen cycloaddition reaction involves a selective cycloaddition reaction (see, *e.g.*, Padwa, A., in COMPREHENSIVE ORGANIC SYNTHESIS, Vol. 4, (ed. Trost, B. M., 1991), p. 1069-1109; Huisgen, R. in 1,3-DIPOLAR CYCLOADDITION CHEMISTRY, (ed. Padwa, A., 1984), p. 1-176) rather than a nucleophilic substitution, the

incorporation of non-naturally encoded amino acids bearing azide and alkyne-containing side chains permits the resultant polypeptides to be modified selectively at the position of the non-naturally encoded amino acid. Cycloaddition reaction involving azide or alkyne-containing antibody can be carried out at room temperature under aqueous conditions by the addition of Cu(II) (including but not limited to, in the form of a catalytic amount of $CuSO_4$) in the presence of a reducing agent for reducing Cu(II) to Cu(I), in situ, in catalytic amount. See, *e.g.*, Wang, Q., et al., J. Am. Chem. Soc. 125, 3192-3193 (2003); Tornoe, C. W., et al., J. Org. Chem. 67:3057-3064 (2002); Rostovtsev, et al., Angew. Chem. Int. Ed. 41:2596-2599 (2002). Exemplary reducing agents include, including but not limited to, ascorbate, metallic copper, quinine, hydroquinone, vitamin K, glutathione, cysteine, Fe^{2+}, Co^{2+}, and an applied electric potential.

[00428] In some cases, where a Huisgen [3+2] cycloaddition reaction between an azide and an alkyne is desired, the antigen-binding polypeptide comprises a non-naturally encoded amino acid comprising an alkyne moiety and the water soluble polymer to be attached to the amino acid comprises an azide moiety. Alternatively, the converse reaction (*i.e.*, with the azide moiety on the amino acid and the alkyne moiety present on the water soluble polymer) can also be performed.

[00429] The azide functional group can also be reacted selectively with a water soluble polymer containing an aryl ester and appropriately functionalized with an aryl phosphine moiety to generate an amide linkage. The aryl phosphine group reduces the azide in situ and the resulting amine then reacts efficiently with a proximal ester linkage to generate the corresponding amide. See, *e.g.*, E. Saxon and C. Bertozzi, Science 287, 2007-2010 (2000). The azide-containing amino acid can be either an alkyl azide (including but not limited to, 2-amino-6-azido-1-hexanoic acid) or an aryl azide (p-azido-phenylalanine).

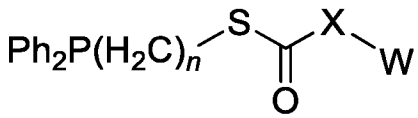
[00430] Exemplary water soluble polymers containing an aryl ester and a phosphine moiety can be represented as follows:



wherein X can be O, N, S or not present, Ph is phenyl, W is a water soluble polymer and R can be H, alkyl, aryl, substituted alkyl and substituted aryl groups. Exemplary R groups include but

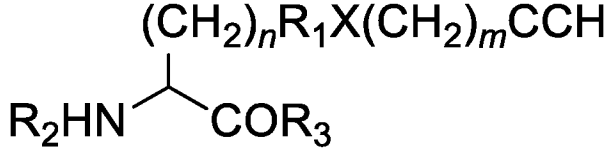
are not limited to $-CH_2$, $-C(CH_3)_3$, –OR', –NR'R", –SR', -halogen, –C(O)R', —CONR'R", – $S(O)_2R'$, $-S(O)_2NR'R"$, –CN and $-NO_2$. R', R", R'" and R"" each independently refer to hydrogen, substituted or unsubstituted heteroalkyl, substituted or unsubstituted aryl, including but not limited to, aryl substituted with 1-3 halogens, substituted or unsubstituted alkyl, alkoxy or thioalkoxy groups, or arylalkyl groups. When a compound of the invention includes more than one R group, for example, each of the R groups is independently selected as are each R', R", R'" and R"" groups when more than one of these groups is present. When R' and R" are attached to the same nitrogen atom, they can be combined with the nitrogen atom to form a 5-, 6-, or 7-membered ring. For example, –NR'R" is meant to include, but not be limited to, 1-pyrrolidinyl and 4-morpholinyl. From the above discussion of substituents, one of skill in the art will understand that the term "alkyl" is meant to include groups including carbon atoms bound to groups other than hydrogen groups, such as haloalkyl (including but not limited to, – CF_3 and $-CH_2CF_3$) and acyl (including but not limited to, —C(O)CH_3, $-C(O)CF_3$, – $C(O)CH_2OCH_3$, and the like).

[00431] The azide functional group can also be reacted selectively with a water soluble polymer containing a thioester and appropriately functionalized with an aryl phosphine moiety to generate an amide linkage. The aryl phosphine group reduces the azide in situ and the resulting amine then reacts efficiently with the thioester linkage to generate the corresponding amide. Exemplary water soluble polymers containing a thioester and a phosphine moiety can be represented as follows:



wherein n is 1-10; X can be O, N, S or not present, Ph is phenyl, and W is a water soluble polymer.

[00432] Exemplary alkyne-containing amino acids can be represented as follows:

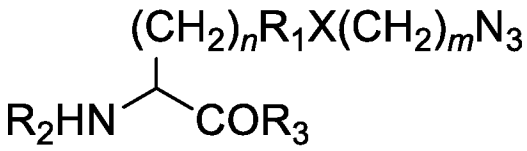


wherein n is 0-10; R_1 is an alkyl, aryl, substituted alkyl, or substituted aryl or not present; X is O, N, S or not present; m is 0-10, R_2 is H, an amino acid, a polypeptide, or an amino terminus modification group, and R_3 is H, an amino acid, a polypeptide, or a carboxy terminus

modification group. In some embodiments, n is 1, R_1 is phenyl, X is not present, m is 0 and the acetylene moiety is positioned in the para position relative to the alkyl side chain. In some embodiments, n is 1, R_1 is phenyl, X is O, m is 1 and the propargyloxy group is positioned in the para position relative to the alkyl side chain (*i.e.*, O-propargyl-tyrosine). In some embodiments, n is 1, R_1 and X are not present and m is 0 (*i.e.*, proparylglycine).

[00433] Alkyne-containing amino acids are commercially available. For example, propargylglycine is commercially available from Peptech (Burlington, Mass.). Alternatively, alkyne-containing amino acids can be prepared according to standard methods. For instance, p-propargyloxyphenylalanine can be synthesized, for example, as described in Deiters, A., et al., J. Am. Chem. Soc. 125: 11782-11783 (2003), and 4-alkynyl-L-phenylalanine can be synthesized as described in Kayser, B., et al., Tetrahedron 53(7): 2475-2484 (1997). Other alkyne-containing amino acids can be prepared by one skilled in the art.

[00434] Exemplary azide-containing amino acids can be represented as follows:



wherein n is 0-10; R_1 is an alkyl, aryl, substituted alkyl, substituted aryl or not present; X is O, N, S or not present; m is 0-10; R_2 is H, an amino acid, a polypeptide, or an amino terminus modification group, and R_3 is H, an amino acid, a polypeptide, or a carboxy terminus modification group. In some embodiments, n is 1, R_1 is phenyl, X is not present, m is 0 and the azide moiety is positioned para to the alkyl side chain. In some embodiments, n is 0-4 and R_1 and X are not present, and m=0. In some embodiments, n is 1, R_1 is phenyl, X is O, m is 2 and the P-azidoethoxy moiety is positioned in the para position relative to the alkyl side chain.

[00435] Azide-containing amino acids are available from commercial sources. For instance, 4-azidophenylalanine can be obtained from Chem-Impex International, Inc. (Wood Dale, Ill.). For those azide-containing amino acids that are not commercially available, the azide group can be prepared relatively readily using standard methods known to those of skill in the art, including but not limited to, via displacement of a suitable leaving group (including but not limited to, halide, mesylate, tosylate) or via opening of a suitably protected lactone. See, *e.g.*, Advanced Organic Chemistry by March (Third Edition, 1985, Wiley and Sons, New York).

[00436] The unique reactivity of beta-substituted aminothiol functional groups makes them extremely useful for the selective modification of polypeptides and other biological molecules that contain aldehyde groups via formation of the thiazolidine. See, *e.g.*, J. Shao and J. Tam, J. Am. Chem. Soc. 1995, 117 (14) 3893-3899. In some embodiments, beta-substituted aminothiol amino acids can be incorporated into antibodies and then reacted with water soluble polymers comprising an aldehyde functionality. In some embodiments, a water soluble polymer, drug conjugate or other payload can be coupled to an antibody polypeptide comprising a beta-substituted aminothiol amino acid via formation of the thiazolidine.

[00437] Particular examples of useful non-natural amino acids include, but are not limited to, p-acetyl-L-phenylalanine, O-methyl-L-tyrosine, L-3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, tri-O-acetyl-GlcNAc b-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-azido-methyl-L-phenylalanine, p-azido-L-phenylalanine, p-acyl-L-phenylalanine, p-benzoyl-L-phenylalanine, L-phosphoserine, phosphonoserine, phosphonotyrosine, p-iodo-phenylalanine, p-bromophenylalanine, p-amino-L-phenylalanine, isopropyl-L-phenylalanine, and p-propargyloxy-phenylalanine. Further useful examples include N-acetyl-L-glucosaminyl-L-serine, N-acetyl-L-galactosaminyl-L-serine, N-acetyl-L-glucosaminyl-L-threonine, N-acetyl-L-glucosaminyl-L-asparagine and O-mannosaminyl-L-serine.

[00438] In particular embodiments, the non-natural amino acids are selected from p-acetyl-phenylalanine, p-ethynyl-phenylalanine, p-propargyloxyphenylalanine, p-azido-methyl-phenylalanine, and p-azido-phenylalanine. One particularly useful non-natural amino acid is p-azido phenylalanine. This amino acid residue is known to those of skill in the art to facilitate Huisgen [3+2] cyloaddition reactions (so-called "click" chemistry reactions) with, for example, compounds bearing alkynyl groups. This reaction enables one of skill in the art to readily and rapidly conjugate to the antibody at the site-specific location of the non-natural amino acid.

[00439] In certain embodiments, the first reactive group is an alkynyl moiety (including but not limited to, in the unnatural amino acid p-propargyloxyphenylalanine, where the propargyl group is also sometimes referred to as an acetylene moiety) and the second reactive group is an azido moiety, and [3+2] cycloaddition chemistry can be used. In certain embodiments, the first reactive group is the azido moiety (including but not limited to, in the unnatural amino acid p-azido-L-phenylalanine) and the second reactive group is the alkynyl moiety.

[00440] In the above formulas, each L represents a divalent linker. The divalent linker can be any divalent linker known to those of skill in the art. Generally, the divalent linker is capable of forming covalent bonds to the functional moiety R and the cognate reactive group (*e.g.*, alpha carbon) of the non-natural amino acid. Useful divalent linkers a bond, alkylene, substituted alkylene, heteroalkylene, substituted heteroalkylene, arylene, substituted arylene, heteroarlyene and substituted heteroarylene. In certain embodiments, L is C_{1-10} alkylene or C_{1-10} heteroalkylene.

[00441] The non-natural amino acids used in the methods and compositions described herein have at least one of the following four properties: (1) at least one functional group on the sidechain of the non-natural amino acid has at least one characteristics and/or activity and/or reactivity orthogonal to the chemical reactivity of the 20 common, genetically-encoded amino acids (*i.e.*, alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, and valine), or at least orthogonal to the chemical reactivity of the naturally occurring amino acids present in the polypeptide that includes the non-natural amino acid; (2) the introduced non-natural amino acids are substantially chemically inert toward the 20 common, genetically-encoded amino acids; (3) the non-natural amino acid can be stably incorporated into a polypeptide, preferably with the stability commensurate with the naturally-occurring amino acids or under typical physiological conditions, and further preferably such incorporation can occur via an in vivo system; and (4) the non-natural amino acid includes an oxime functional group or a functional group that can be transformed into an oxime group by reacting with a reagent, preferably under conditions that do not destroy the biological properties of the polypeptide that includes the non-natural amino acid (unless of course such a destruction of biological properties is the purpose of the modification/transformation), or where the transformation can occur under aqueous conditions at a pH between about 4 and about 8, or where the reactive site on the non-natural amino acid is an electrophilic site. Any number of non-natural amino acids can be introduced into the polypeptide. Non-natural amino acids may also include protected or masked oximes or protected or masked groups that can be transformed into an oxime group after deprotection of the protected group or unmasking of the masked group. Non-natural amino acids may also include protected or masked carbonyl or dicarbonyl groups, which can be transformed into a carbonyl or dicarbonyl group after deprotection of the protected group or unmasking of the

masked group and thereby are available to react with hydroxylamines or oximes to form oxime groups.

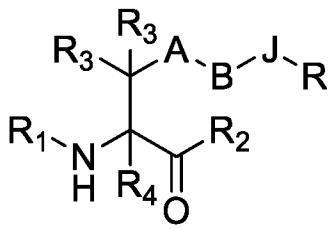
[00442] In further embodiments, non-natural amino acids that may be used in the methods and compositions described herein include, but are not limited to, amino acids comprising a photoactivatable cross-linker, spin-labeled amino acids, fluorescent amino acids, metal binding amino acids, metal-containing amino acids, radioactive amino acids, amino acids with novel functional groups, amino acids that covalently or non-covalently interact with other molecules, photocaged and/or photoisomerizable amino acids, amino acids comprising biotin or a biotin analogue, glycosylated amino acids such as a sugar substituted serine, other carbohydrate modified amino acids, keto-containing amino acids, aldehyde-containing amino acids, amino acids comprising polyethylene glycol or other polyethers, heavy atom substituted amino acids, chemically cleavable and/or photocleavable amino acids, amino acids with an elongated side chains as compared to natural amino acids, including but not limited to, polyethers or long chain hydrocarbons, including but not limited to, greater than about 5 or greater than about 10 carbons, carbon-linked sugar-containing amino acids, redox-active amino acids, amino thioacid containing amino acids, and amino acids comprising one or more toxic moiety.

[00443] In some embodiments, non-natural amino acids comprise a saccharide moiety. Examples of such amino acids include N-acetyl-L-glucosaminyl-L-serine, N-acetyl-L-galactosaminyl-L-serine, N-acetyl-L-glucosaminyl-L-threonine, N-acetyl-L-glucosaminyl-L-asparagine and O-mannosaminyl-L-serine. Examples of such amino acids also include examples where the naturally-occurring N- or O-linkage between the amino acid and the saccharide is replaced by a covalent linkage not commonly found in nature–including but not limited to, an alkene, an oxime, a thioether, an amide and the like. Examples of such amino acids also include saccharides that are not commonly found in naturally-occurring proteins such as 2-deoxy-glucose, 2-deoxygalactose and the like.

[00444] The chemical moieties incorporated into antibodies via incorporation of non-natural amino acids offer a variety of advantages and manipulations of polypeptides. For example, the unique reactivity of a carbonyl or dicarbonyl functional group (including a keto- or aldehyde-functional group) allows selective modification of antibodies with any of a number of hydrazine- or hydroxylamine-containing reagents in vivo and in vitro. A heavy atom non-natural amino acid, for example, can be useful for phasing x-ray structure data. The site-specific introduction of heavy atoms using non-natural amino acids also provides selectivity and

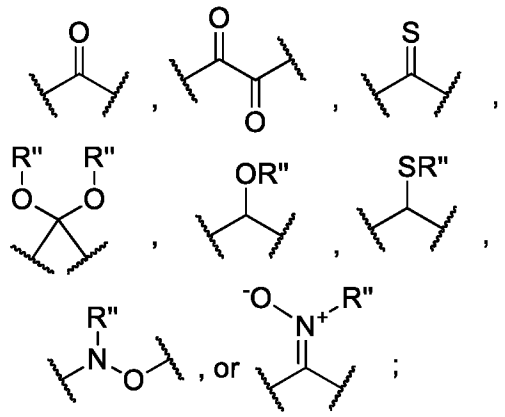
flexibility in choosing positions for heavy atoms. Photoreactive non-natural amino acids (including but not limited to, amino acids with benzophenone and arylazides (including but not limited to, phenylazide) side chains), for example, allow for efficient in vivo and in vitro photocrosslinking of polypeptides. Examples of photoreactive non-natural amino acids include, but are not limited to, p-azido-phenylalanine and p-benzoyl-phenylalanine. The antibodies with the photoreactive non-natural amino acids may then be crosslinked at will by excitation of the photoreactive group-providing temporal control. In a non-limiting example, the methyl group of a non-natural amino can be substituted with an isotopically labeled, including but not limited to, with a methyl group, as a probe of local structure and dynamics, including but not limited to, with the use of nuclear magnetic resonance and vibrational spectroscopy.

[00445] Amino acids with an electrophilic reactive group allow for a variety of reactions to link molecules via various chemical reactions, including, but not limited to, nucleophilic addition reactions. Such electrophilic reactive groups include a carbonyl- or dicarbonyl-group (including a keto- or aldehyde group), a carbonyl-like- or dicarbonyl-like-group (which has reactivity similar to a carbonyl- or dicarbonyl-group and is structurally similar to a carbonyl- or dicarbonyl-group), a masked carbonyl- or masked dicarbonyl-group (which can be readily converted into a carbonyl- or dicarbonyl-group), or a protected carbonyl- or protected dicarbonyl-group (which has reactivity similar to a carbonyl- or dicarbonyl-group upon deprotection). Such amino acids include amino acids having the structure of Formula (I):



wherein: A is optional, and when present is lower alkylene, substituted lower alkylene, lower cycloalkylene, substituted lower cycloalkylene, lower alkenylene, substituted lower alkenylene, alkynylene, lower heteroalkylene, substituted heteroalkylene, lower heterocycloalkylene, substituted lower heterocycloalkylene, arylene, substituted arylene, heteroarylene, substituted heteroarylene, alkarylene, substituted alkarylene, aralkylene, or substituted aralkylene; B is optional, and when present is a linker selected from the group consisting of lower alkylene, substituted lower alkylene, lower alkenylene, substituted lower alkenylene, lower heteroalkylene, substituted lower heteroalkylene, –O–, –O–(alkylene or

substituted alkylene)–, –S–, –S–(alkylene or substituted alkylene)–, –S(O)$_k$– where k is 1, 2, or 3, –S(O)$_k$(alkylene or substituted alkylene)–, –C(O)–, –NS(O)$_2$–, –OS(O)$_2$–, –C(O)–(alkylene or substituted alkylene)–, –C(S)–, –C(S)–(alkylene or substituted alkylene)–, —N(R')–, –NR'–(alkylene or substituted alkylene)–, —C(O)N(R')–, –CON(R')–(alkylene or substituted alkylene)–, –CSN(R')–, –CSN(R')–(alkylene or substituted alkylene)–, —N(R')CO–(alkylene or substituted alkylene)–, –N(R')C(O)O–, —S(O)$_k$N(R')–, —N(R')C(O)N(R')–, –N(R')C(S)N(R')–, –N(R')S(O)$_k$N(R')–, –N(R')–N=, —C(R')=N–, —C(R')=N–N(R')–, –C(R')=N–N=, –C(R')$_2$–N=N–, and –C(R')$_2$–N(R')–N(R')–, where each R' is independently H, alkyl, or substituted alkyl; J is



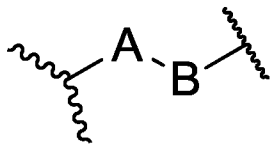
R is H, alkyl, substituted alkyl, cycloalkyl, or substituted cycloalkyl; each R" is independently H, alkyl, substituted alkyl, or a protecting group, or when more than one R" group is present, two R" optionally form a heterocycloalkyl; R_1 is H, an amino protecting group, resin, amino acid, polypeptide, or polynucleotide; and R_2 is OH, an ester protecting group, resin, amino acid, polypeptide, or polynucleotide; each of R_3 and R_4 is independently H, halogen, lower alkyl, or substituted lower alkyl, or R_3 and R_4 or two R_3 groups optionally form a cycloalkyl or a heterocycloalkyl; or the -A-B-J-R groups together form a bicyclic or tricyclic cycloalkyl or heterocycloalkyl comprising at least one carbonyl group, including a dicarbonyl group, protected carbonyl group, including a protected dicarbonyl group, or masked carbonyl group, including a masked dicarbonyl group; or the -J-R group together forms a monocyclic or bicyclic cycloalkyl or heterocycloalkyl comprising at least one carbonyl group, including a dicarbonyl group, protected carbonyl group, including a protected dicarbonyl group, or masked carbonyl group, including a masked dicarbonyl group; with a proviso that when A is phenylene and each R_3 is H, B is present; and that when A is –$(CH_2)_4$– and each R_3 is H, B is not –NHC(O)(CH_2CH_2)–; and that when A and B are absent and each R_3 is H, R is not methyl. Such non-natural amino acids may be in the form of a salt, or may be incorporated into a non-natural

amino acid polypeptide, polymer, polysaccharide, or a polynucleotide and optionally post translationally modified.

[00446] In certain embodiments, compounds of Formula (I) are stable in aqueous solution for at least 1 month under mildly acidic conditions. In certain embodiments, compounds of Formula (I) are stable for at least 2 weeks under mildly acidic conditions. In certain embodiments, compound of Formula (I) are stable for at least 5 days under mildly acidic conditions. In certain embodiments, such acidic conditions are pH 2 to 8.

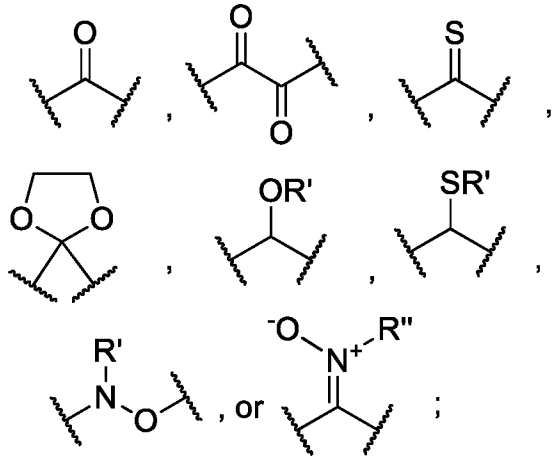
[00447] In certain embodiments of compounds of Formula (I), B is lower alkylene, substituted lower alkylene, –O–(alkylene or substituted alkylene)–, –C(R')=N–N(R')–, —N(R')CO–, –C(O)-, –C(R')=N–, –C(O)–(alkylene or substituted alkylene)–, –CON(R')–(alkylene or substituted alkylene)–, –S(alkylene or substituted alkylene)–, –S(O)(alkylene or substituted alkylene)–, or —$S(O)_2$(alkylene or substituted alkylene)–. In certain embodiments of compounds of Formula (I), B is –$O(CH_2)$–, –CH=N–, –CH=N–NH–, –$NHCH_2$–, —NHCO–, –C(O)–, –C(O)–(CH_2)–, —CONH–(CH_2)–, –SCH_2–, –S(=O)CH_2–, or —$S(O)_2CH_2$–. In certain embodiments of compounds of Formula (I), R is C_1–6 alkyl or cycloalkyl. In certain embodiments of compounds of Formula (I) R is –CH_3, –$CH(CH_3)_2$, or cyclopropyl. In certain embodiments of compounds of Formula (I), R_1 is H, tert-butyloxycarbonyl (Boc), 9-Fluorenylmethoxycarbonyl (Fmoc), N-acetyl, tetrafluoroacetyl (TFA), or benzyloxycarbonyl (Cbz). In certain embodiments of compounds of Formula (I), R_1 is a resin, amino acid, polypeptide, or polynucleotide. In certain embodiments of compounds of Formula (I), R_2 is OH, O-methyl, O-ethyl, or O-t-butyl. In certain embodiments of compounds of Formula (I), R_2 is a resin, amino acid, polypeptide, or polynucleotide. In certain embodiments of compounds of Formula (I), R_2 is a polynucleotide. In certain embodiments of compounds of Formula (I), R_2 is ribonucleic acid (RNA). In certain embodiments of compounds of Formula (I), R_2 is tRNA. In certain embodiments of compounds of Formula (I), the tRNA specifically recognizes a selector codon. In certain embodiments of compounds of Formula (I) the selector codon is selected from the group consisting of an amber codon, ochre codon, opal codon, a unique codon, a rare codon, an unnatural codon, a five-base codon, and a four-base codon. In certain embodiments of compounds of Formula (I), R_2 is a suppressor tRNA.

[00448] In certain embodiments of compounds of Formula (I),



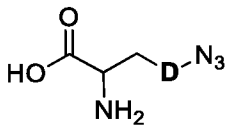
is selected from the group consisting of: (i) A is substituted lower alkylene, C_4-arylene, substituted arylene, heteroarylene, substituted heteroarylene, alkarylene, substituted alkarylene, aralkylene, or substituted aralkylene; B is optional, and when present is a divalent linker selected from the group consisting of lower alkylene, substituted lower alkylene, lower alkenylene, substituted lower alkenylene, –O–, –O–(alkylene or substituted alkylene)–, –S–, –S(O)–, $-S(O)_2-$, $-NS(O)_2-$, $-OS(O)_2-$, –C(O)–, –C(O)–(alkylene or substituted alkylene)–, –C(S)–, –N(R')–, –C(O)N(R')–, –CON(R')–(alkylene or substituted alkylene)–, –CSN(R')–, —N(R')CO–(alkylene or substituted alkylene)–, –N(R')C(O)O–, –N(R')C(S)–, –S(O)N(R'), $-S(O)_2N(R')$, –N(R')C(O)N(R')–, —N(R')C(S)N(R')–, –N(R')S(O)N(R')–, $-N(R')S(O)_2N(R')-$, –N(R')–N=, –C(R')=N–N(R')–, —C(R')=N–N=, $-C(R')_2-N=N-$, and $-C(R')_2-N(R')-N(R')-$; (ii) A is optional, and when present is substituted lower alkylene, C_4–arylene, substituted arylene, heteroarylene, substituted heteroarylene, alkarylene, substituted alkarylene, aralkylene, or substituted aralkylene; B is a divalent linker selected from the group consisting of lower alkylene, substituted lower alkylene, lower alkenylene, substituted lower alkenylene, –O–, –O–(alkylene or substituted alkylene)–, –S-, –S(O)–, $-S(O)_2-$, $-NS(O)_2-$, $-OS(O)_2-$, –C(O)–, –C(O)–(alkylene or substituted alkylene)–, —C(S)–, –N(R')–, –C(O)N(R')–, –CON(R')–(alkylene or substituted alkylene)–, –CSN(R')–, —N(R')CO–(alkylene or substituted alkylene)–, –N(R')C(O)O–, –N(R')C(S)–, –S(O)N(R'), $-S(O)_2N(R')$, –N(R')C(O)N(R')–, –N(R')C(S)N(R')–, –N(R')S(O)N(R')–, $-N(R')S(O)_2N(R')-$, —N(R')–N=, –C(R')=N–N(R')–, –C(R')=N–N=, $-C(R')_2-N=N-$, and $-C(R')_2-N(R')-N(R')-$; (iii) A is lower alkylene; B is optional, and when present is a divalent linker selected from the group consisting of lower alkylene, substituted lower alkylene, lower alkenylene, substituted lower alkenylene, –O–, –O–(alkylene or substituted alkylene)–, –S–, –S(O)–, $-S(O)_2-$, $-NS(O)_2-$, $-OS(O)_2-$, –C(O)–, –C(O)–(alkylene or substituted alkylene)–, –C(S)–, –N(R')–, –C(O)N(R')–, –CSN(R')–, –CON(R')–(alkylene or substituted alkylene)–, –N(R')C(O)O–, –N(R')C(S)–, —S(O)N(R'), $-S(O)_2N(R')$, —N(R')C(O)N(R')–, –N(R')C(S)N(R')–, –N(R')S(O)N(R')–, $-N(R')S(O)_2N(R')-$, –N(R')–N=, –C(R')=N–N(R')–, –C(R')=N–N=, $-C(R')_2-N=N-$, and $-C(R')_2-N(R')-N(R')-$; and (iv) A is phenylene; B is a divalent linker selected from the group consisting of lower alkylene, substituted lower alkylene, lower alkenylene, substituted lower alkenylene, –O–, —O-(alkylene or substituted alkylene)–, –S–,

–S(O)–, –S(O)$_2$–, –NS(O)$_2$–, —OS(O)$_2$–, —C(O)–, —C(O)–(alkylene or substituted alkylene)–, –C(S)–, –N(R')–, –C(O)N(R')-, —CON(R')–(alkylene or substituted alkylene)–, –CSN(R')–, –N(R')CO–(alkylene or substituted alkylene)–, –N(R')C(O)O–, –N(R')C(S)–, –S(O)N(R'), –S(O)$_2$N(R'), —N(R')C(O)N(R')–, —N(R')C(S)N(R')–, –N(R')S(O)N(R')–, –N(R')S(O)$_2$N(R')–, –N(R')–N=, –C(R')'N–N(R')–, —C(R')=N–N=, –C(R')$_2$–N=N–, and –C(R')$_2$–N(R')–N(R')–; J is



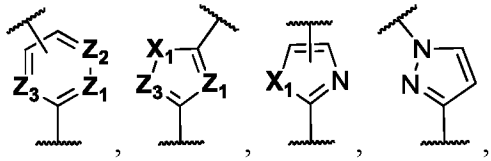
each R' is independently H, alkyl, or substituted alkyl; R_1 is optional, and when present, is H, an amino protecting group, resin, amino acid, polypeptide, or polynucleotide; and R_2 is optional, and when present, is OH, an ester protecting group, resin, amino acid, polypeptide, or polynucleotide; and each R_3 and R_4 is independently H, halogen, lower alkyl, or substituted lower alkyl; and R is H, alkyl, substituted alkyl, cycloalkyl, or substituted cycloalkyl.

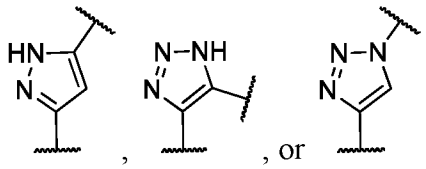
[00449] In certain embodiments, the non-natural amino acid can be according to formula XIX:



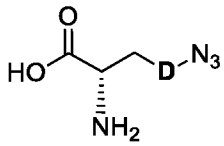
Formula XIX;

or a salt thereof, wherein: **D** is –**Ar**-**W$_3$**– or –**W$_1$**-**Y$_1$**-C(O)-**Y$_2$**-**W$_2$**–; **Ar** is



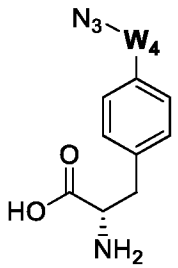


each of W_1, W_2, and W_3 is independently a single bond or lower alkylene; each X_1 is independently –NH–, –O–, or –S–; each Y_1 is independently a single bond, –NH–, or –O–; each Y_2 is independently a single bond, –NH–, –O–, or an N-linked or C-linked pyrrolidinylene; and one of Z_1, Z_2, and Z_3 is –N– and the others of Z_1, Z_2, and Z_3 are independently –CH–. In certain embodiments, the non-natural amino acid is according to formula XIXa:



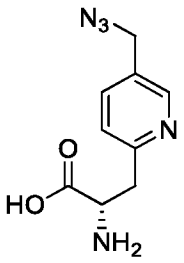
Formula XIXa;

where **D** is a defined in the context of formula XIX. In certain embodiments, the non-natural amino acid is according formula XIXb:

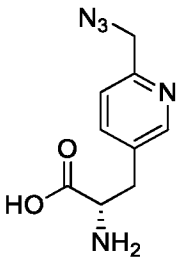


Formula XIXb;

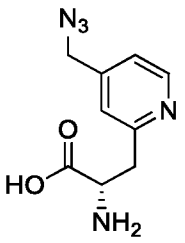
or a salt thereof, wherein W_4 is C_1-C_{10} alkylene. In a further embodiment, W_4 is C_1-C_5 alkylene. In an embodiment, W_4 is C_1-C_3 alkylene. In an embodiment, W_4 is C_1 alkylene. In particular embodiments, the non-natural amino acid is selected from the group consisting of:



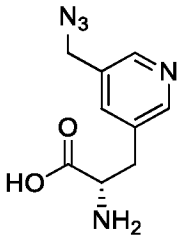
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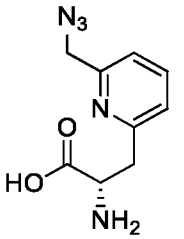
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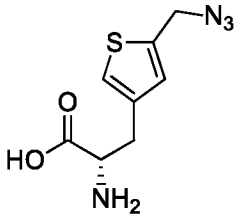
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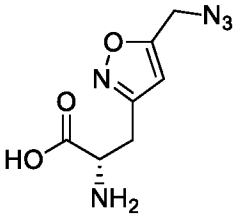
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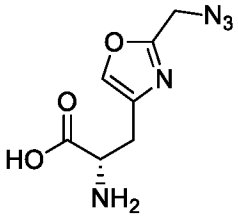
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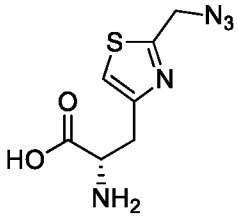
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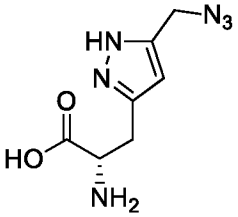
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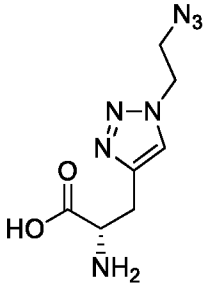
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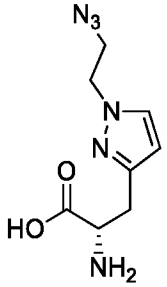
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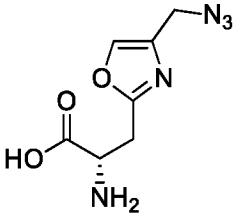
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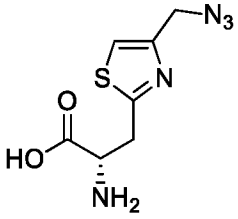
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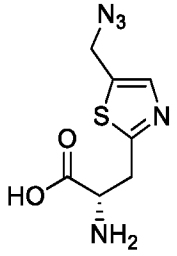
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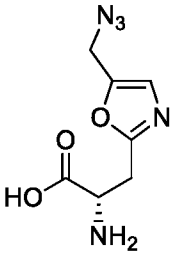
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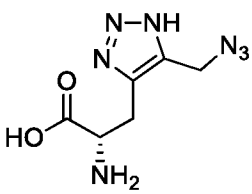
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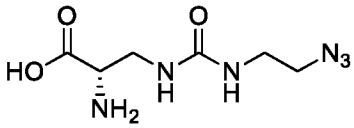
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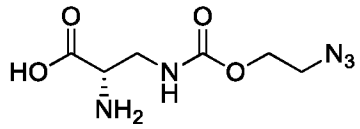
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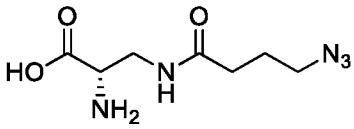
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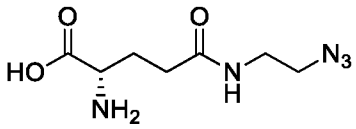
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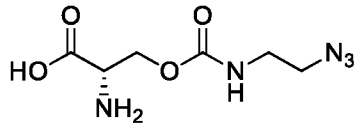
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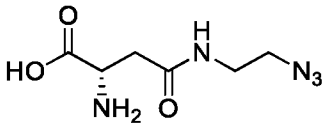
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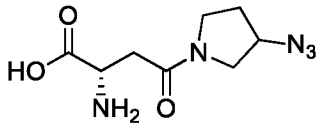
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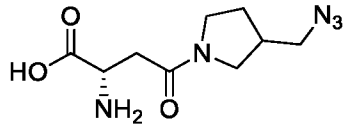
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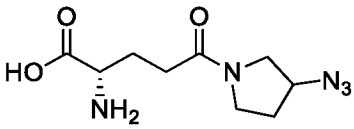
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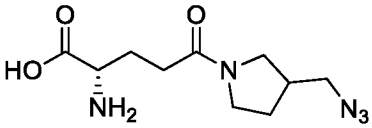
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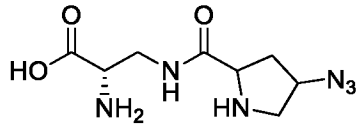
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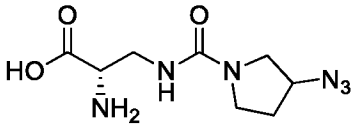
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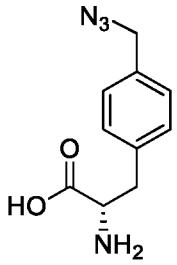
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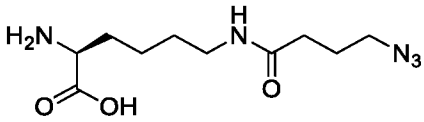
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(29)



(30);

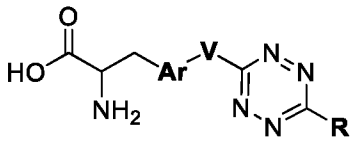


(40);

and

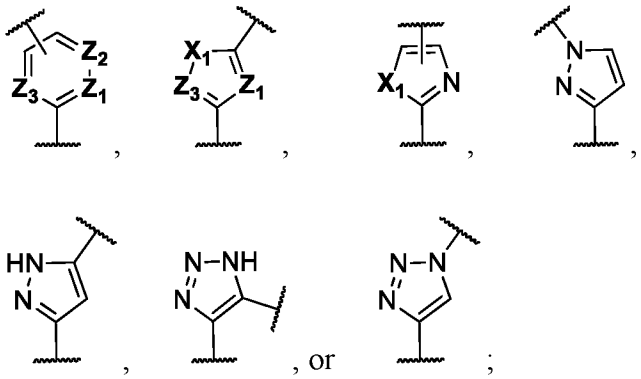
or a salt thereof. Such non-natural amino acids may be in the form of a salt, or may be incorporated into a non-natural amino acid polypeptide, polymer, polysaccharide, or a polynucleotide and optionally post translationally modified.

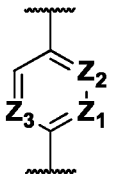
[00450] In certain embodiments, the modified amino acid is according to formula I:



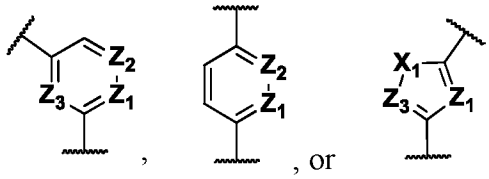
Formula I;

or a salt thereof, wherein **Ar** is:



V is a single bond, lower alkylene, or –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or lower alkylene, and the other is –NH–, –O–, or –S–; each $\mathbf{X_1}$ is independently –NH–, –O–, or –S–; one of $\mathbf{Z_1}$, $\mathbf{Z_2}$, and $\mathbf{Z_3}$ is –CH– or –N– and the others of $\mathbf{Z_1}$, $\mathbf{Z_2}$, and $\mathbf{Z_3}$ are each independently –CH–; and **R** is lower alkyl. In certain embodiments, when **Ar** is  and **V** is –NH–, then one of $\mathbf{Z_1}$, $\mathbf{Z_2}$, and $\mathbf{Z_3}$ is –N–. In certain embodiments, **V** is a single bond, –NH–, **or** —CH_2NH–.

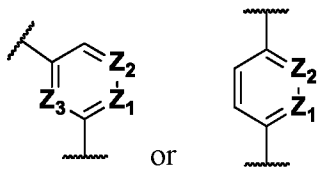
[00451] In certain embodiments, **Ar** is



and $\mathbf{Z_1}$, $\mathbf{Z_2}$, $\mathbf{Z_3}$ and $\mathbf{X_1}$ are as defined in the context of formula I. In certain embodiments according to this paragraph, **V** is –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments according to this paragraph, **V** is a single bond, –NH–, **or** —CH_2NH–. In certain embodiments according to this paragraph, $\mathbf{Z_1}$ is N. In certain embodiments according to this paragraph, $\mathbf{Z_2}$ is N. In certain embodiments according

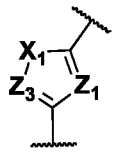
to this paragraph, Z_3 is N. In certain embodiments according to this paragraph, Z_1 is CH, Z_3 is CH and X_1 is S.

[00452] In certain embodiments, **Ar** is



and Z_1, Z_2, and Z_3 are as defined in the context of formula I. In certain embodiments according to this paragraph, **V** is $-W_1-W_2-$; one of W_1 and W_2 is absent or $-CH_2-$, and the other is –NH–, —O–, or –S–. In certain embodiments according to this paragraph, **V** is a single bond, –NH–, or $-CH_2NH-$. In certain embodiments according to this paragraph, Z_1 is N. In certain embodiments according to this paragraph, Z_2 is N. In certain embodiments according to this paragraph, Z_3 is N.

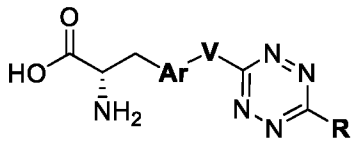
[00453] In certain embodiments, **Ar** is



;

and Z_1, Z_3 and X_1 are as defined in the context of formula I. In certain embodiments according to this paragraph, **V** is $-W_1-W_2-$; one of W_1 and W_2 is absent or $-CH_2-$, and the other is –NH–, –O–, or –S–. In certain embodiments according to this paragraph, **V** is a single bond, –NH–, or $-CH_2NH-$. In certain embodiments according to this paragraph, Z_1 is N. In certain embodiments according to this paragraph, Z_3 is N. In certain embodiments according to this paragraph, Z_1 is CH, Z_3 is CH and X_1 is S.

[00454] In certain embodiments, the modified amino acid is according to Formula Ia:

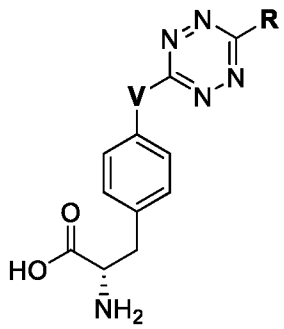


Formula Ia;

where **Ar**, **V**, and **R** are defined in the context of formula I.

[00455] In an embodiment, compounds of either of formulas I and Ia are provided wherein **V** is a single bond. In another embodiment, compounds of either of formulas I and Ia are provided wherein **V** is –NH–. In another embodiment, compounds of either of formulas I and Ia are provided wherein **V** is $–CH_2NH–$.

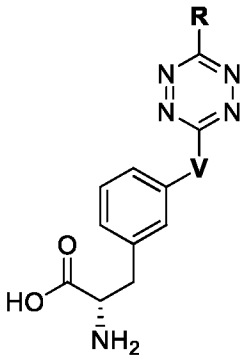
[00456] In certain embodiments, the modified amino acid is according to Formula II:



Formula II;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is $–W_1–W_2–$; one of W_1 and W_2 is absent or $–CH_2–$, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or $—CH_2NH–$. In certain embodiments, **V** is a single bond or $–CH_2NH–$; and **R** is methyl.

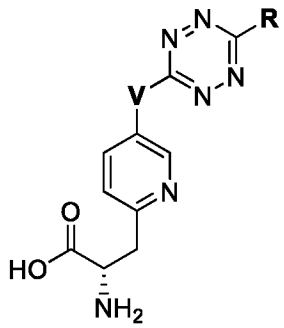
[00457] In certain embodiments, the modified amino acid is according to Formula III:



Formula III;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is $–W_1–W_2–$; one of W_1 and W_2 is absent or $–CH_2–$, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or $—CH_2NH–$. In certain embodiments, **V** is a single bond, –NH–, or $–CH_2NH–$; and **R** is methyl.

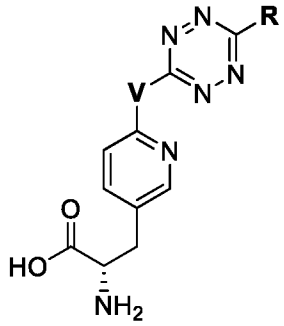
[00458] In certain embodiments, the modified amino acid is according to Formula IV:



Formula IV;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

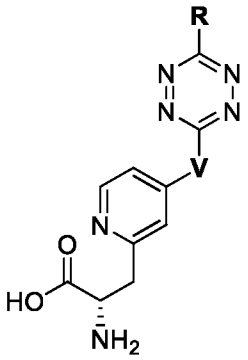
[00459] In certain embodiments, the modified amino acid is according to Formula V:



Formula V;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

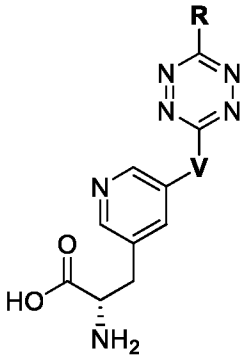
[00460] In certain embodiments, the modified amino acid is according to Formula VI:



Formula VI;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

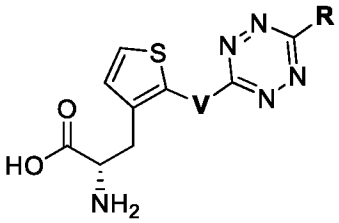
[00461] In certain embodiments, the modified amino acid is according to Formula VII:



Formula VII;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –$\mathbf{W_1}$–$\mathbf{W_2}$–; one of $\mathbf{W_1}$ and $\mathbf{W_2}$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

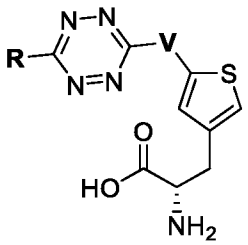
[00462] In certain embodiments, the modified amino acid is according to Formula VIII:



Formula VIII;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –**W**$_1$–**W**$_2$–; one of **W**$_1$ and **W**$_2$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

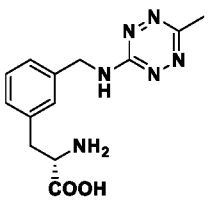
[00463] In certain embodiments, the modified amino acid is according to Formula IX:



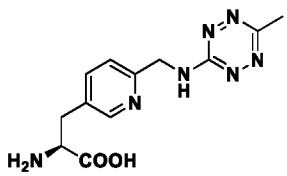
Formula IX;

or a salt thereof, wherein **V** and **R** are as defined in Formula I. In certain embodiments according to this paragraph, **V** is –**W**$_1$–**W**$_2$–; one of **W**$_1$ and **W**$_2$ is absent or –CH_2–, and the other is –NH–, –O–, or –S–. In certain embodiments, **V** is a single bond, –NH–, or —CH_2NH–. In certain embodiments, **V** is a single bond, –NH–, or –CH_2NH–; and **R** is methyl.

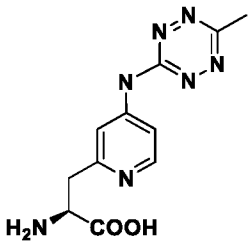
[00464] In certain embodiments, the modified amino acid is according to any of formulas 51-62:



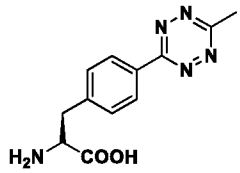
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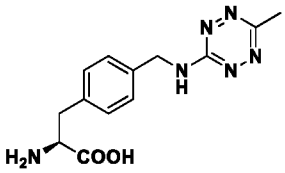
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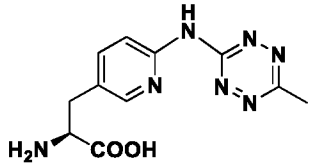
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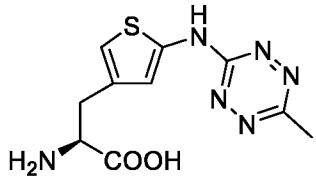
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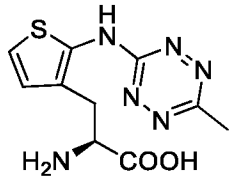
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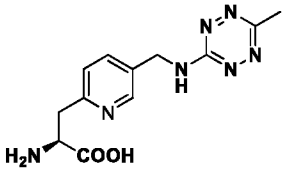
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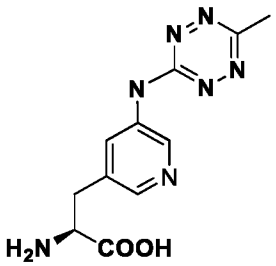
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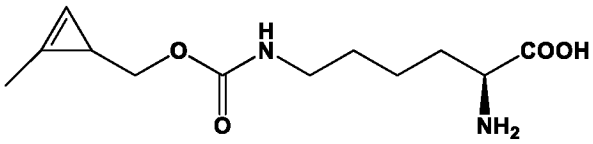


(58)

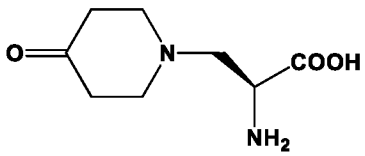


(59)





(60) (61)



(62)

or a salt thereof.

[00465] In certain embodiments, the non-natural amino acid is selected from the group consisting of compounds 30, 53, 56, 59, 60, 61, and 62 above. In certain embodiments, the non-natural amino acid is compound 30. In certain embodiments, the non-natural amino acid is compound 56. In some embodiments, the non-natural amino acid is compound 61. In some embodiments, the non-natural amino acid is compound 62.

12. Preparation of Antibody Conjugates

12.1. Antigen Preparation

[00466] The FOLR1 protein to be used for isolation of the antibodies may be intact FOLR1 or a fragment of FOLR1. The intact FOLR1 protein, or fragment of FOLR1, may be in the form of an isolated protein or protein expressed by a cell. Other forms of FOLR1 useful for generating antibodies will be apparent to those skilled in the art.

12.2. Monoclonal Antibodies

[00467] Monoclonal antibodies may be obtained, for example, using the hybridoma method first described by Kohler et al., *Nature*, 1975, 256:495-497 (incorporated by reference in its entirety), and/or by recombinant DNA methods (*see e.g.,* U.S. Patent No. 4,816,567, incorporated by reference in its entirety). Monoclonal antibodies may also be obtained, for example, using phage or yeast-based libraries. *See e.g.,* U.S. Patent Nos. 8,258,082 and 8,691,730, each of which is incorporated by reference in its entirety.

[00468] In the hybridoma method, a mouse or other appropriate host animal is immunized to elicit lymphocytes that produce or are capable of producing antibodies that will specifically bind to the protein used for immunization. Alternatively, lymphocytes may be immunized *in vitro*. Lymphocytes are then fused with myeloma cells using a suitable fusing agent, such as polyethylene glycol, to form a hybridoma cell. *See* Goding J.W., *Monoclonal Antibodies: Principles and Practice* 3rd ed. (1986) Academic Press, San Diego, CA, incorporated by reference in its entirety.

[00469] The hybridoma cells are seeded and grown in a suitable culture medium that contains one or more substances that inhibit the growth or survival of the unfused, parental myeloma cells. For example, if the parental myeloma cells lack the enzyme hypoxanthine guanine phosphoribosyl transferase (HGPRT or HPRT), the culture medium for the hybridomas typically will include hypoxanthine, aminopterin, and thymidine (HAT medium), which substances prevent the growth of HGPRT-deficient cells.

[00470] Useful myeloma cells are those that fuse efficiently, support stable high-level production of antibody by the selected antibody-producing cells, and are sensitive media conditions, such as the presence or absence of HAT medium. Among these, preferred myeloma cell lines are murine myeloma lines, such as those derived from MOP-21 and MC-11 mouse tumors (available from the Salk Institute Cell Distribution Center, San Diego, CA), and SP-2

or X63-Ag8-653 cells (available from the American Type Culture Collection, Rockville, MD). Human myeloma and mouse-human heteromyeloma cell lines also have been described for the production of human monoclonal antibodies. *See e.g.,* Kozbor, *J. Immunol.*, 1984, 133:3001, incorporated by reference in its entirety.

[00471] After the identification of hybridoma cells that produce antibodies of the desired specificity, affinity, and/or biological activity, selected clones may be subcloned by limiting dilution procedures and grown by standard methods. *See* Goding, *supra*. Suitable culture media for this purpose include, for example, D-MEM or RPMI-1640 medium. In addition, the hybridoma cells may be grown *in vivo* as ascites tumors in an animal.

[00472] DNA encoding the monoclonal antibodies may be readily isolated and sequenced using conventional procedures (e.g., by using oligonucleotide probes that are capable of binding specifically to genes encoding the heavy and light chains of the monoclonal antibodies). Thus, the hybridoma cells can serve as a useful source of DNA encoding antibodies with the desired properties. Once isolated, the DNA may be placed into expression vectors, which are then transfected into host cells such as bacteria (e.g., *E. coli*), yeast (e.g., *Saccharomyces* or *Pichia* sp.), COS cells, Chinese hamster ovary (CHO) cells, or myeloma cells that do not otherwise produce antibody, to produce the monoclonal antibodies.

12.3. Humanized Antibodies

[00473] Humanized antibodies may be generated by replacing most, or all, of the structural portions of a non-human monoclonal antibody with corresponding human antibody sequences. Consequently, a hybrid molecule is generated in which only the antigen-specific variable, or CDR, is composed of non-human sequence. Methods to obtain humanized antibodies include those described in, for example, Winter and Milstein, *Nature*, 1991, 349:293-299; Rader et al., *Proc. Nat. Acad. Sci. U.S.A.*, 1998, 95:8910-8915; Steinberger et al., *J. Biol. Chem.*, 2000, 275:36073-36078; Queen et al., *Proc. Natl. Acad. Sci. U.S.A.*, 1989, 86:10029-10033; and U.S. Patent Nos. 5,585,089, 5,693,761, 5,693,762, and 6,180,370; each of which is incorporated by reference in its entirety.

12.4. Human Antibodies

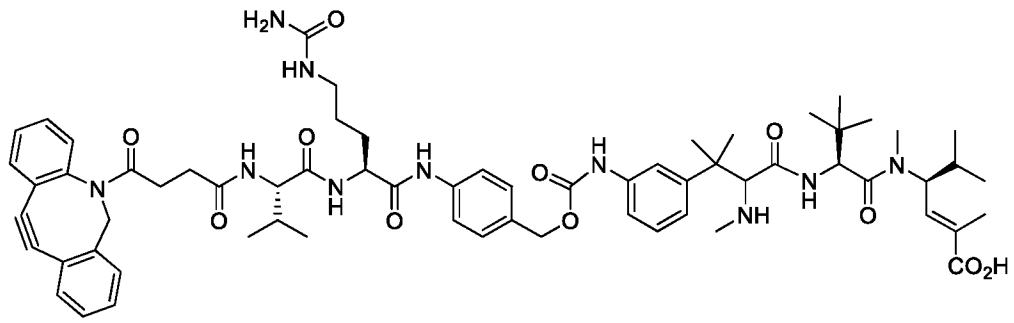
[00474] Human antibodies can be generated by a variety of techniques known in the art, for example by using transgenic animals (e.g., humanized mice). *See, e.g.,* Jakobovits et al., *Proc. Natl. Acad. Sci. U.S.A.*, 1993, 90:2551; Jakobovits et al., *Nature*, 1993, 362:255-258; Bruggermann et al., *Year in Immuno.*, 1993, 7:33; and U.S. Patent Nos. 5,591,669, 5,589,369

and 5,545,807; each of which is incorporated by reference in its entirety. Human antibodies can also be derived from phage-display libraries (*see e.g.,* Hoogenboom et al., *J. Mol. Biol.*, 1991, 227:381-388; Marks et al., *J. Mol. Biol.*, 1991, 222:581-597; and U.S. Pat. Nos. 5,565,332 and 5,573,905; each of which is incorporated by reference in its entirety). Human antibodies may also be generated by *in vitro* activated B cells (*see e.g.,* U.S. Patent. Nos. 5,567,610 and 5,229,275, each of which is incorporated by reference in its entirety). Human antibodies may also be derived from yeast-based libraries (*see e.g.,* U.S. Patent No. 8,691,730, incorporated by reference in its entirety).

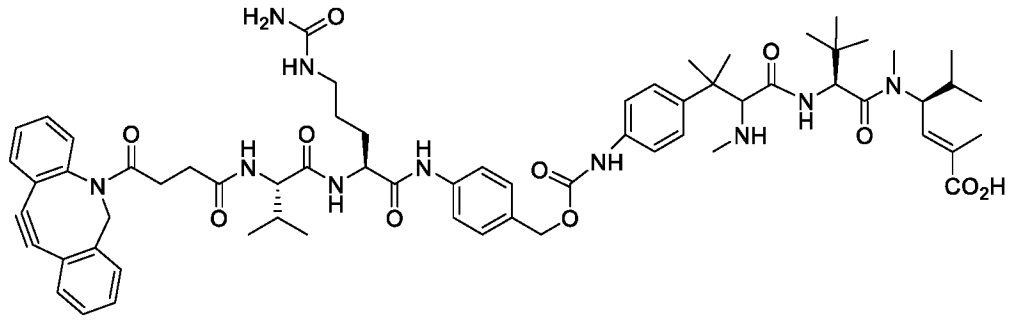
12.5. Conjugation

[00475] The antibody conjugates can be prepared by standard techniques. In certain embodiments, an antibody is contacted with a payload precursor under conditions suitable for forming a bond from the antibody to the payload to form an antibody-payload conjugate. In certain embodiments, an antibody is contacted with a linker precursor under conditions suitable for forming a bond from the antibody to the linker. The resulting antibody-linker is contacted with a payload precursor under conditions suitable for forming a bond from the antibody-linker to the payload to form an antibody-linker-payload conjugate. In certain embodiments, a payload precursor is contacted with a linker precursor under conditions suitable for forming a bond from the payload to the linker. The resulting payload-linker is contacted with an antibody under conditions suitable for forming a bond from the payload-linker to the antibody to form an antibody-linker-payload conjugate. Suitable linkers for preparing the antibody conjugates are disclosed herein, and exemplary conditions for conjugation are described in the Examples below.

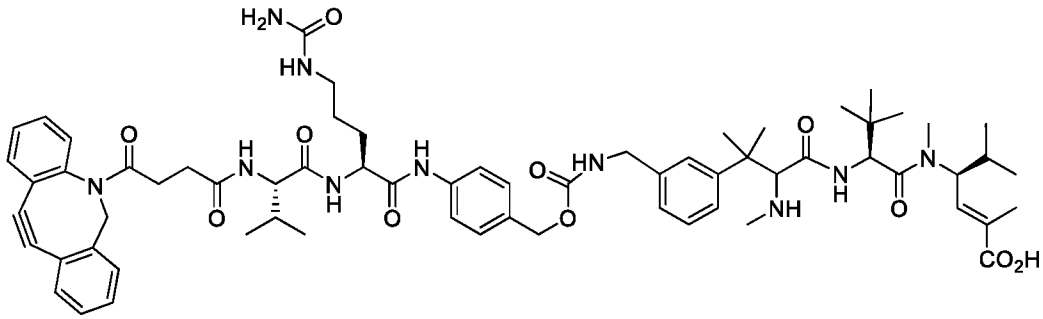
[00476] In some embodiments, an anti-FOLR1 conjugate is prepared by contacting an anti-FOLR1 antibody as disclosed herein with a linker precursor having a structure of any of (A) – (L):



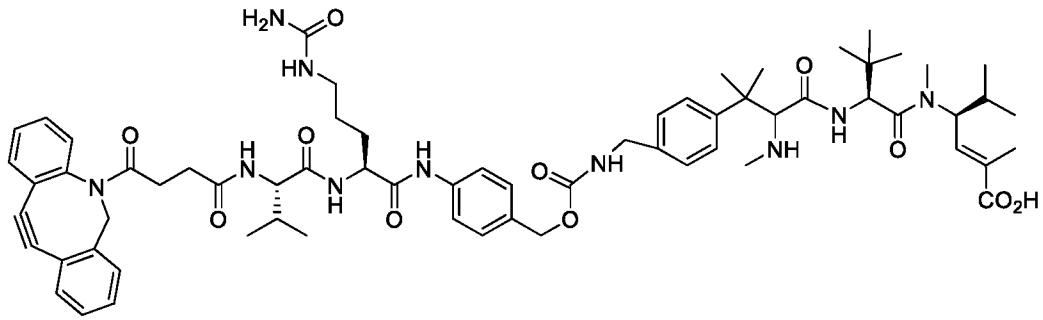
(A)



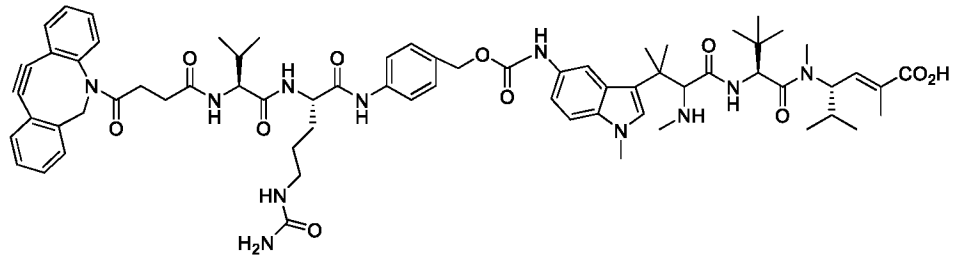
(B)



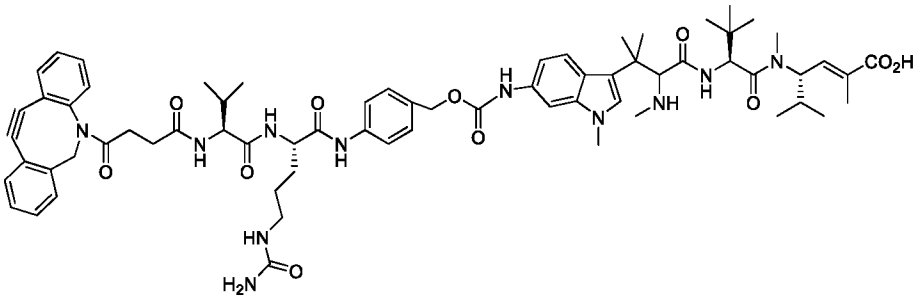
(C)



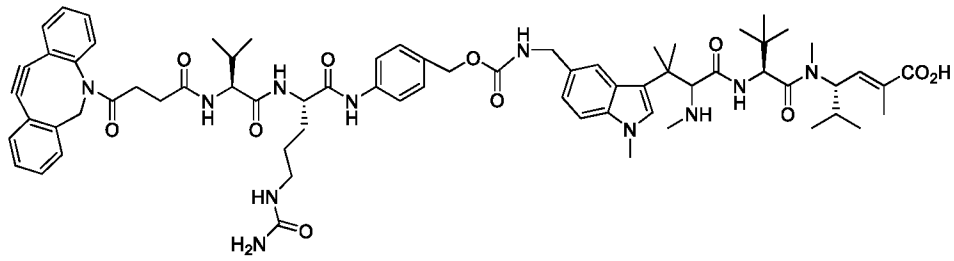
(D)



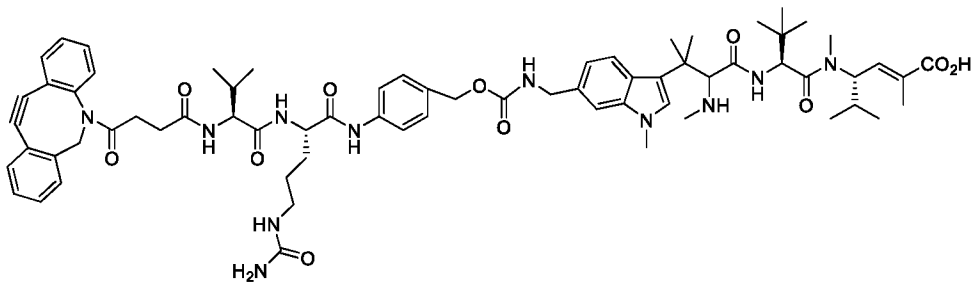
(E)



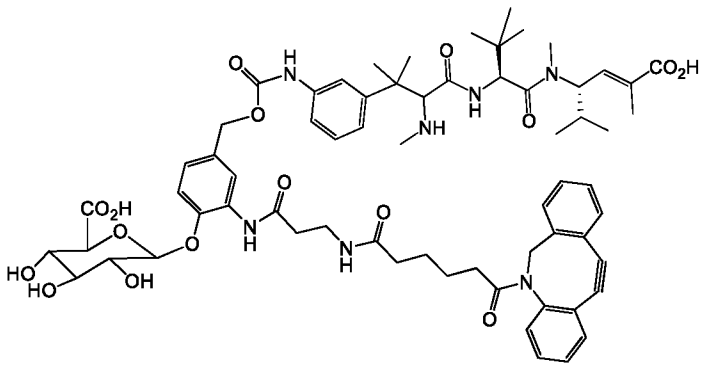
(F)



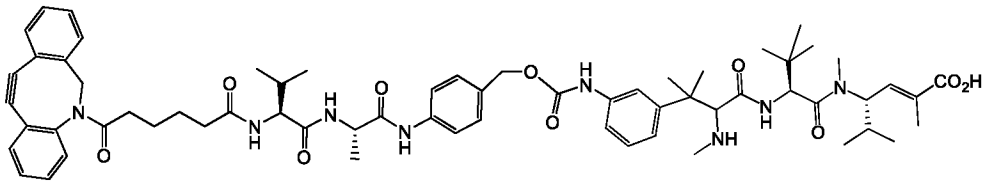
(G)



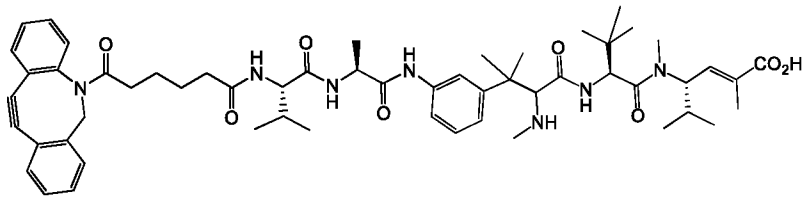
(H)



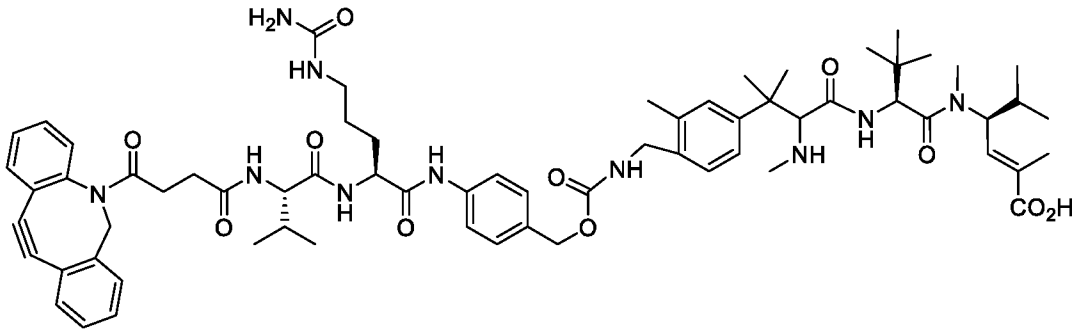
(I)



(J)

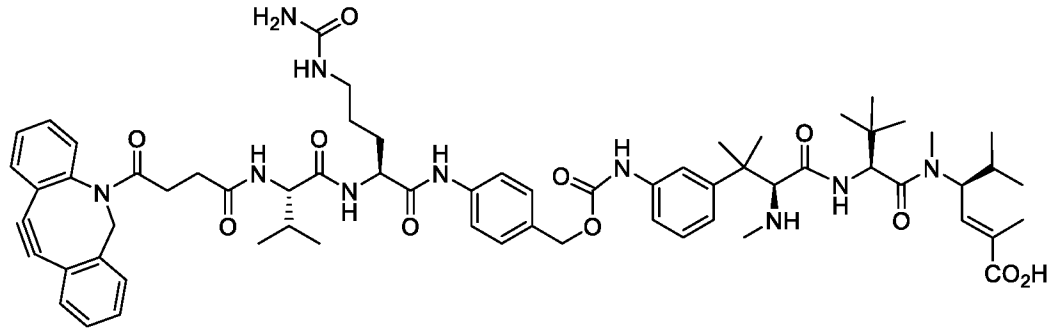


(K)

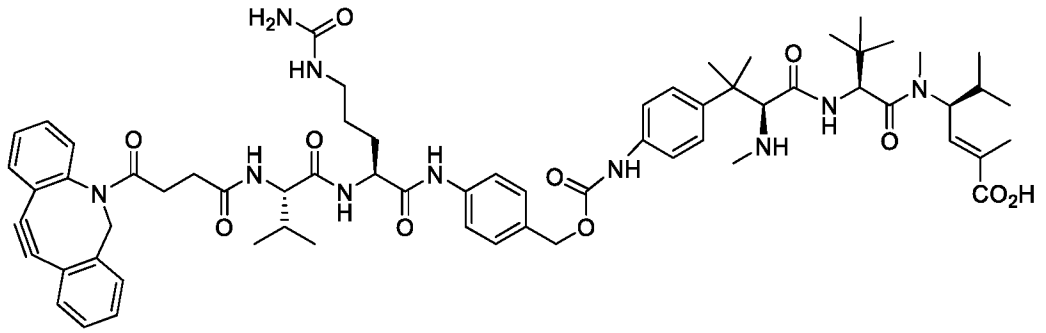


(L)

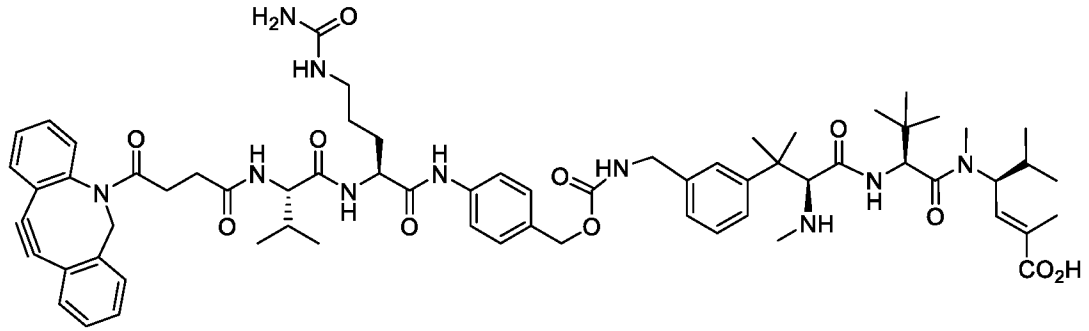
[00477] In some embodiments, the stereochemistry of the linker precursors identified as (A) – (L) is identified with **R** and **S** notation for each chiral center, from left to right as depicted in formulas (A1) – (L1) and (A2) – (L2) illustrated below:



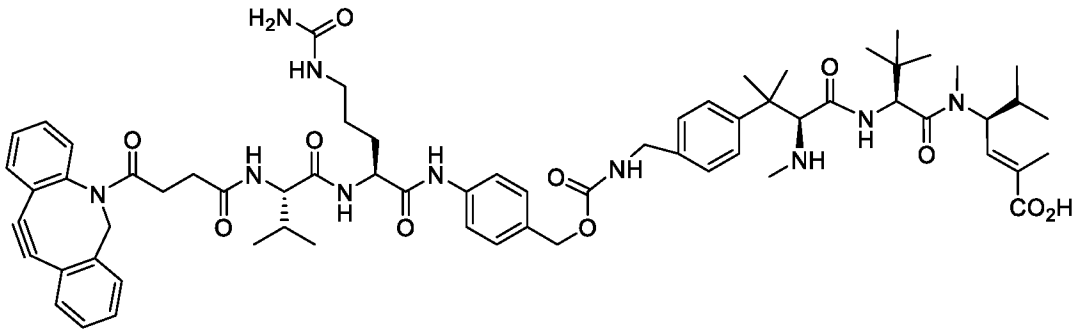
(A1)



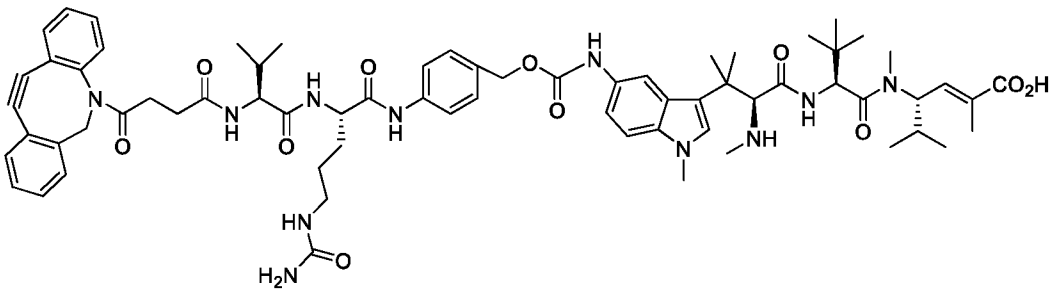
(B1)



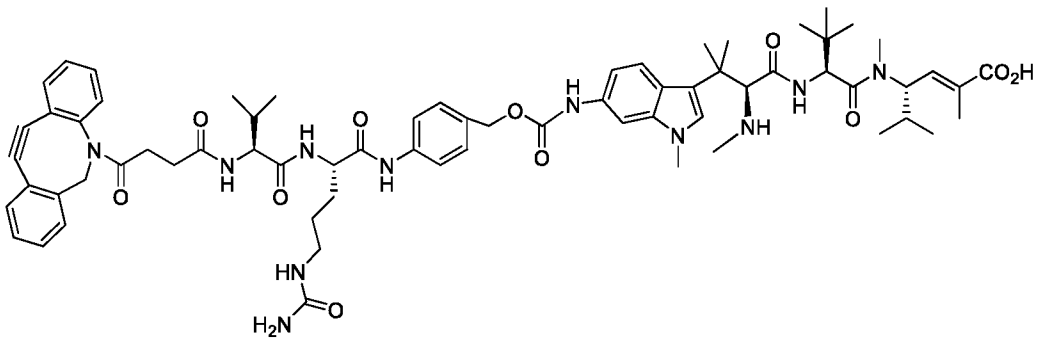
(C1)



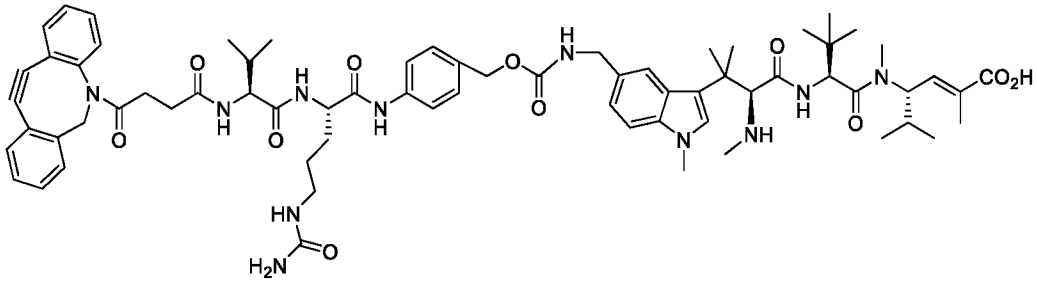
(D1)



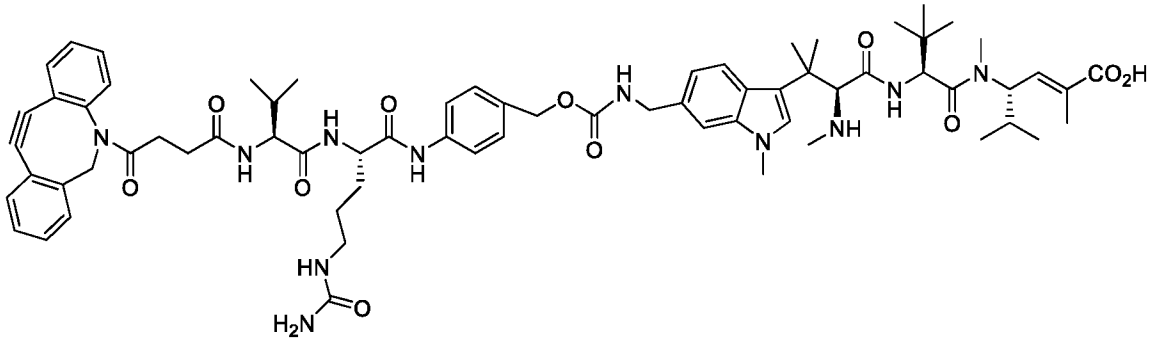
(E1)



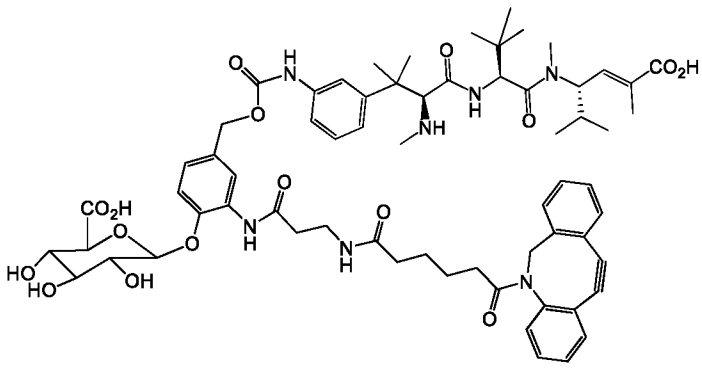
(F1)



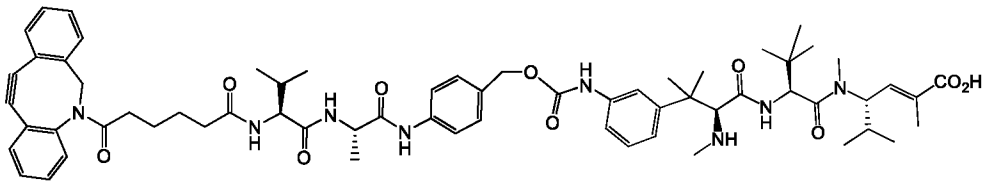
(G1)



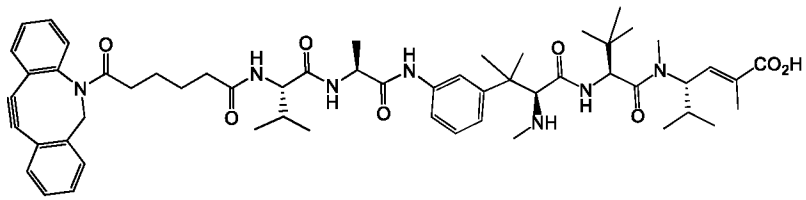
(H1)



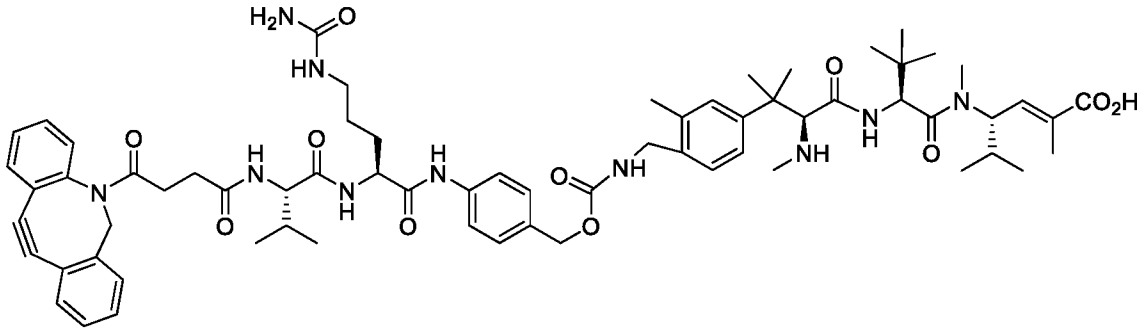
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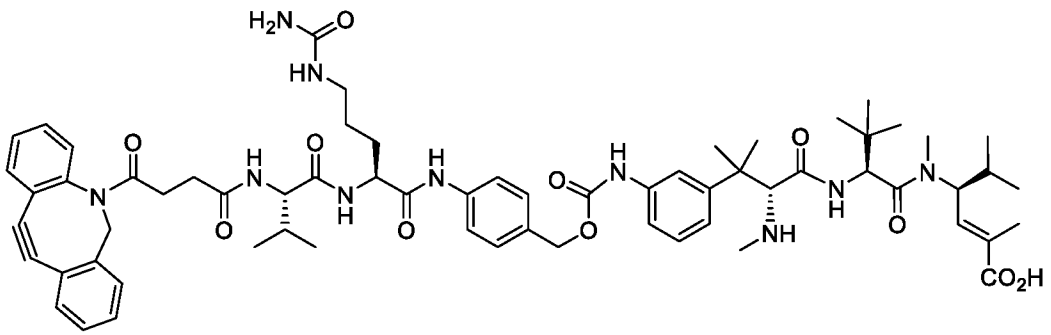
(J1)



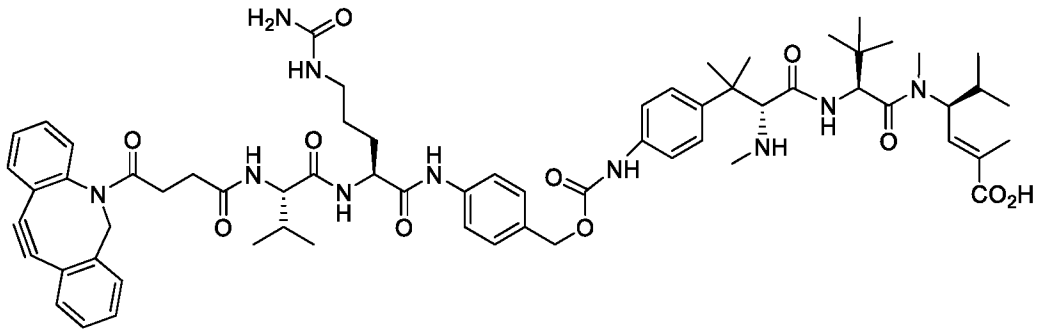
(K1)



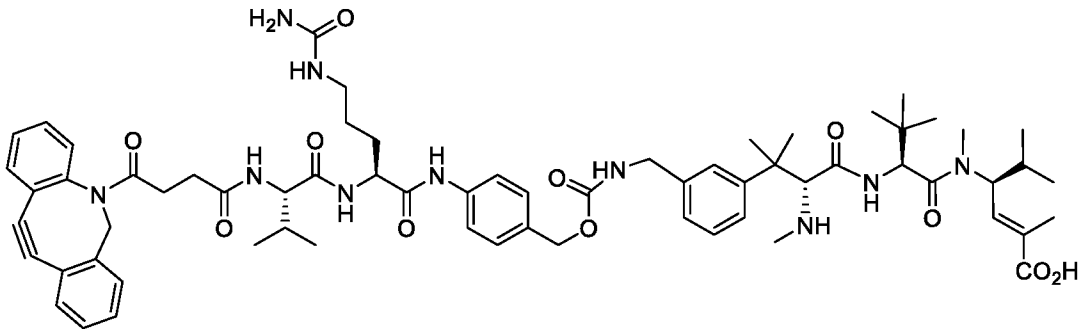
(L1)



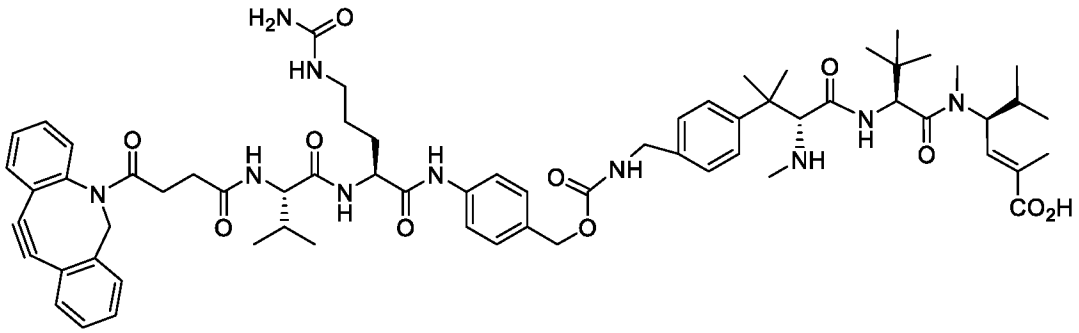
(A2)



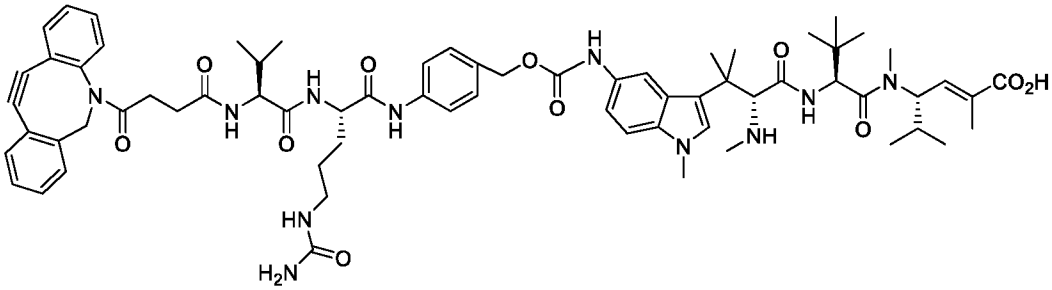
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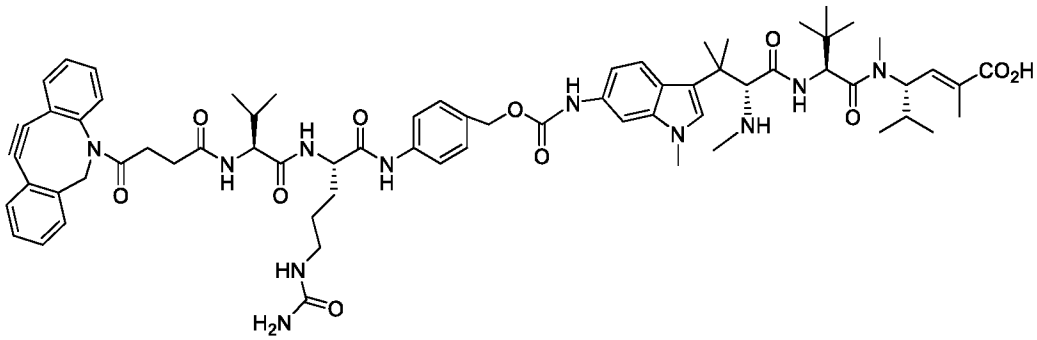
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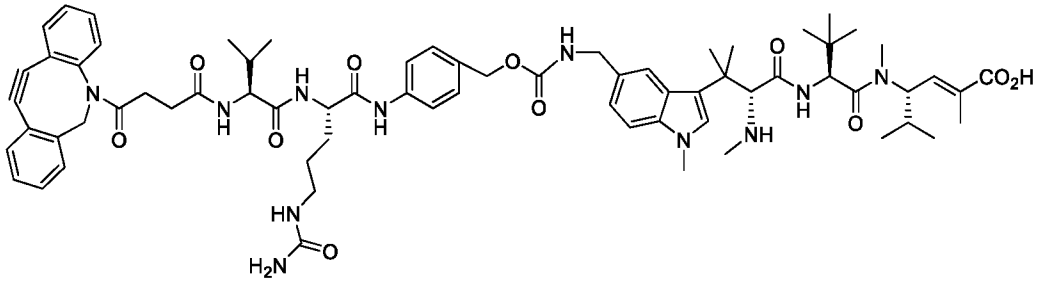
(D2)



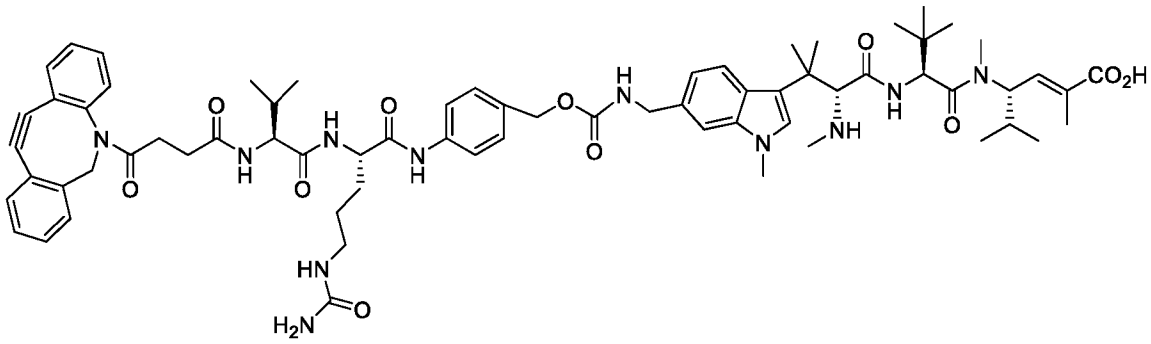
(E2)



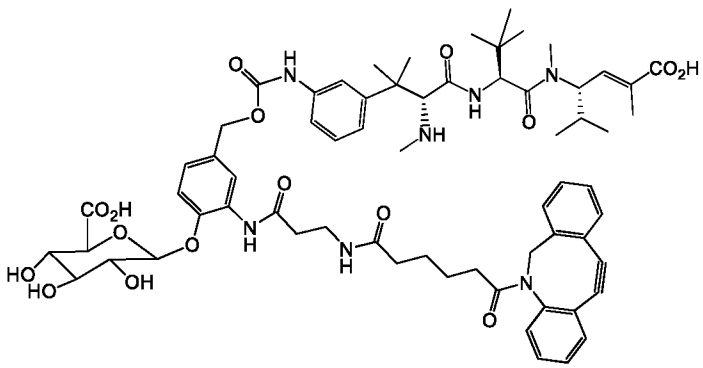
(F2)



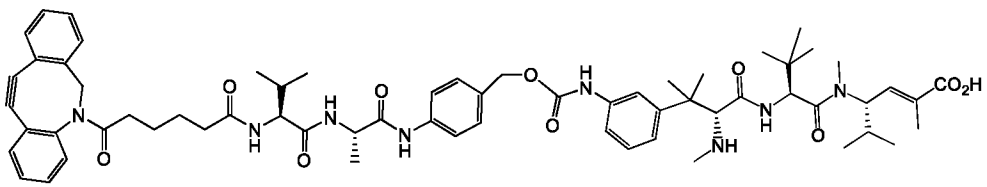
(G2)



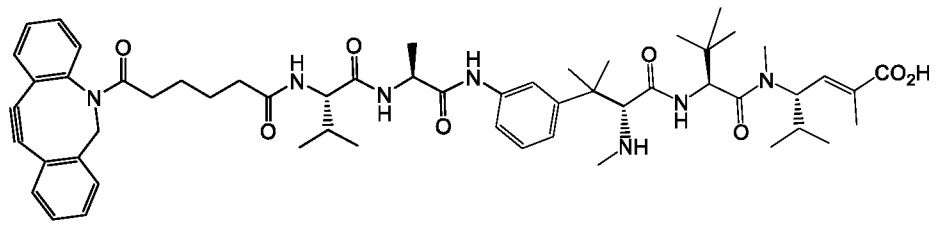
(H2)



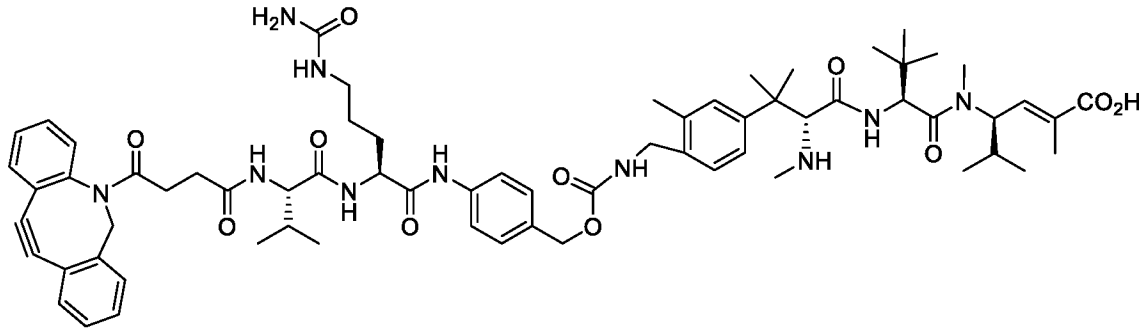
(I2)



(J2)



(K2)



(L2)

13. Vectors, Host Cells, and Recombinant Methods

[00478] Embodiments are also directed to the provision of isolated nucleic acids encoding anti-FOLR1 antibodies, vectors and host cells comprising the nucleic acids, and recombinant techniques for the production of the antibodies.

[00479] For recombinant production of the antibody, the nucleic acid(s) encoding it may be isolated and inserted into a replicable vector for further cloning (i.e., amplification of the DNA) or expression. In some aspects, the nucleic acid may be produced by homologous recombination, for example as described in U.S. Patent No. 5,204,244, incorporated by reference in its entirety.

[00480] Many different vectors are known in the art. The vector components generally include, but are not limited to, one or more of the following: a signal sequence, an origin of replication, one or more marker genes, an enhancer element, a promoter, and a transcription termination sequence, for example as described in U.S. Patent No. 5,534,615, incorporated by reference in its entirety.

[00481] Illustrative examples of suitable host cells are provided below. These host cells are not meant to be limiting.

[00482] Suitable host cells include any prokaryotic (e.g., bacterial), lower eukaryotic (e.g., yeast), or higher eukaryotic (e.g., mammalian) cells. Suitable prokaryotes include eubacteria, such as Gram-negative or Gram-positive organisms, for example, Enterobacteriaceae such as *Escherichia* (*E. coli*), *Enterobacter*, *Erwinia*, *Klebsiella*, *Proteus*, *Salmonella* (*S. typhimurium*), *Serratia* (*S. marcescans*), *Shigella*, *Bacilli* (*B. subtilis* and *B. licheniformis*), *Pseudomonas* (*P. aeruginosa*), and *Streptomyces*. One useful *E. coli* cloning host is *E. coli* 294, although other strains such as *E. coli* B, *E. coli* X1776, and *E. coli* W3110 are suitable.

[00483] In addition to prokaryotes, eukaryotic microbes such as filamentous fungi or yeast are also suitable cloning or expression hosts for anti-FOLR1 antibody-encoding vectors. *Saccharomyces cerevisiae*, or common baker's yeast, is a commonly used lower eukaryotic host microorganism. However, a number of other genera, species, and strains are available and useful, such as *Spodoptera frugiperda* (*e.g.*, SF9), *Schizosaccharomyces pombe*, *Kluyveromyces* (*K. lactis*, *K. fragilis*, *K. bulgaricus K. wickeramii*, *K. waltii*, *K. drosophilarum*, *K. thermotolerans*, and *K. marxianus*), *Yarrowia*, *Pichia pastoris*, *Candida* (*C. albicans*), *Trichoderma reesia*, *Neurospora crassa*, *Schwanniomyces* (*S. occidentalis*), and filamentous fungi such as, for example *Penicillium*, *Tolypocladium*, and *Aspergillus* (*A. nidulans* and *A. niger*).

[00484] Useful mammalian host cells include COS-7 cells, HEK293 cells; baby hamster kidney (BHK) cells; Chinese hamster ovary (CHO); mouse sertoli cells; African green monkey kidney cells (VERO-76), and the like.

[00485] The host cells used to produce the anti-FOLR1 antibody of this invention may be cultured in a variety of media. Commercially available media such as, for example, Ham's F10, Minimal Essential Medium (MEM), RPMI-1640, and Dulbecco's Modified Eagle's Medium (DMEM) are suitable for culturing the host cells. In addition, any of the media described in Ham et al., *Meth. Enz.*, 1979, 58:44; Barnes et al., *Anal. Biochem.*, 1980, 102:255; and U.S. Patent Nos. 4,767,704, 4,657,866, 4,927,762, 4,560,655, and 5,122,469, or WO 90/03430 and WO 87/00195 may be used. Each of the foregoing references is incorporated by reference in its entirety.

[00486] Any of these media may be supplemented as necessary with hormones and/or other growth factors (such as insulin, transferrin, or epidermal growth factor), salts (such as sodium chloride, calcium, magnesium, and phosphate), buffers (such as HEPES), nucleotides (such as adenosine and thymidine), antibiotics, trace elements (defined as inorganic compounds usually

present at final concentrations in the micromolar range), and glucose or an equivalent energy source. Any other necessary supplements may also be included at appropriate concentrations that would be known to those skilled in the art.

[00487] The culture conditions, such as temperature, pH, and the like, are those previously used with the host cell selected for expression, and will be apparent to the ordinarily skilled artisan.

[00488] When using recombinant techniques, the antibody can be produced intracellularly, in the periplasmic space, or directly secreted into the medium. If the antibody is produced intracellularly, as a first step, the particulate debris, either host cells or lysed fragments, is removed, for example, by centrifugation or ultrafiltration. For example, Carter et al. (*Bio/Technology*, 1992, 10:163-167) describes a procedure for isolating antibodies which are secreted to the periplasmic space of *E. coli*. Briefly, cell paste is thawed in the presence of sodium acetate (pH 3.5), EDTA, and phenylmethylsulfonylfluoride (PMSF) over about 30 min. Cell debris can be removed by centrifugation.

[00489] In some embodiments, the antibody is produced in a cell-free system. In some aspects, the cell-free system is an *in vitro* transcription and translation system as described in Yin et al., *mAbs*, 2012, 4:217-225, incorporated by reference in its entirety. In some aspects, the cell-free system utilizes a cell-free extract from a eukaryotic cell or from a prokaryotic cell. In some aspects, the prokaryotic cell is *E. coli*. Cell-free expression of the antibody may be useful, for example, where the antibody accumulates in a cell as an insoluble aggregate, or where yields from periplasmic expression are low. The antibodies produced in a cell-free system may be aglycosylated depending on the source of the cells.

[00490] Where the antibody is secreted into the medium, supernatants from such expression systems are generally first concentrated using a commercially available protein concentration filter, for example, an Amicon® or Millipore® Pellcon® ultrafiltration unit. A protease inhibitor such as PMSF may be included in any of the foregoing steps to inhibit proteolysis and antibiotics may be included to prevent the growth of adventitious contaminants.

[00491] The antibody composition prepared from the cells can be purified using, for example, hydroxylapatite chromatography, gel electrophoresis, dialysis, and affinity chromatography, with affinity chromatography being a particularly useful purification technique. The suitability of protein A as an affinity ligand depends on the species and isotype of any immunoglobulin Fc domain that is present in the antibody. Protein A can be used to

purify antibodies that are based on human γ1, γ2, or γ4 heavy chains (Lindmark et al., *J. Immunol. Meth.*, 1983, 62:1-13, incorporated by reference in its entirety). Protein G is useful for all mouse isotypes and for human γ3 (Guss et al., *EMBO J.*, 1986, 5:1567-1575, incorporated by reference in its entirety).

[00492] The matrix to which the affinity ligand is attached is most often agarose, but other matrices are available. Mechanically stable matrices such as controlled pore glass or poly(styrenedivinyl)benzene allow for faster flow rates and shorter processing times than can be achieved with agarose. Where the antibody comprises a C_{H3} domain, the BakerBond ABX® resin is useful for purification.

[00493] Other techniques for protein purification, such as fractionation on an ion-exchange column, ethanol precipitation, Reverse Phase HPLC, chromatography on silica, chromatography on heparin Sepharose®, chromatofocusing, SDS-PAGE, and ammonium sulfate precipitation are also available, and can be applied by one of skill in the art.

[00494] Following any preliminary purification step(s), the mixture comprising the antibody of interest and contaminants may be subjected to low pH hydrophobic interaction chromatography using an elution buffer at a pH between about 2.5 to about 4.5, generally performed at low salt concentrations (e.g., from about 0 to about 0.25 M salt).

14. Pharmaceutical Compositions and Methods of Administration

[00495] The antibody conjugates provided herein can be formulated into pharmaceutical compositions using methods available in the art and those disclosed herein. Any of the antibody conjugates provided herein can be provided in the appropriate pharmaceutical composition and be administered by a suitable route of administration.

[00496] The methods provided herein encompass administering pharmaceutical compositions comprising at least one antibody conjugate provided herein and one or more compatible and pharmaceutically acceptable carriers. In this context, the term "pharmaceutically acceptable" means approved by a regulatory agency of the Federal or a state government or listed in the U.S. Pharmacopeia or other generally recognized pharmacopeia for use in animals, and more particularly in humans. The term "carrier" includes a diluent, adjuvant (*e.g.*, Freund's adjuvant (complete and incomplete)), excipient, or vehicle with which the therapeutic is administered. Such pharmaceutical carriers can be sterile liquids, such as water and oils, including those of petroleum, animal, vegetable or synthetic origin, such as peanut oil, soybean oil, mineral oil, sesame oil and the like. Water can be used as a carrier when the

pharmaceutical composition is administered intravenously. Saline solutions and aqueous dextrose and glycerol solutions can also be employed as liquid carriers, particularly for injectable solutions. Examples of suitable pharmaceutical carriers are described in Martin, E.W., *Remington's Pharmaceutical Sciences*.

[00497] In clinical practice the pharmaceutical compositions or antibody conjugates provided herein may be administered by any route known in the art. Exemplary routes of administration include, but are not limited to, the inhalation, intraarterial, intradermal, intramuscular, intraperitoneal, intravenous, nasal, parenteral, pulmonary, and subcutaneous routes. In some embodiments, a pharmaceutical composition or antibody conjugate provided herein is administered parenterally.

[00498] The compositions for parenteral administration can be emulsions or sterile solutions. Parenteral compositions may include, for example, propylene glycol, polyethylene glycol, vegetable oils, and injectable organic esters (e.g., ethyl oleate). These compositions can also contain wetting, isotonizing, emulsifying, dispersing and stabilizing agents. Sterilization can be carried out in several ways, for example using a bacteriological filter, by radiation or by heating. Parenteral compositions can also be prepared in the form of sterile solid compositions which can be dissolved at the time of use in sterile water or any other injectable sterile medium.

[00499] In some embodiments, a composition provided herein is a pharmaceutical composition or a single unit dosage form. Pharmaceutical compositions and single unit dosage forms provided herein comprise a prophylactically or therapeutically effective amount of one or more prophylactic or therapeutic antibody conjugates.

[00500] The pharmaceutical composition may comprise one or more pharmaceutical excipients. Any suitable pharmaceutical excipient may be used, and one of ordinary skill in the art is capable of selecting suitable pharmaceutical excipients. Non-limiting examples of suitable excipients include starch, glucose, lactose, sucrose, gelatin, malt, rice, flour, chalk, silica gel, sodium stearate, glycerol monostearate, talc, sodium chloride, dried skim milk, glycerol, propylene, glycol, water, ethanol and the like. Whether a particular excipient is suitable for incorporation into a pharmaceutical composition or dosage form depends on a variety of factors well known in the art including, but not limited to, the way in which the dosage form will be administered to a subject and the specific antibody in the dosage form. The composition or single unit dosage form, if desired, can also contain minor amounts of wetting or emulsifying agents, or pH buffering agents. Accordingly, the pharmaceutical

excipients provided below are intended to be illustrative, and not limiting. Additional pharmaceutical excipients include, for example, those described in the *Handbook of Pharmaceutical Excipients*, Rowe et al. (Eds.) 6th Ed. (2009), incorporated by reference in its entirety.

[00501] In some embodiments, the pharmaceutical composition comprises an anti-foaming agent. Any suitable anti-foaming agent may be used. In some aspects, the anti-foaming agent is selected from an alcohol, an ether, an oil, a wax, a silicone, a surfactant, and combinations thereof. In some aspects, the anti-foaming agent is selected from a mineral oil, a vegetable oil, ethylene bis stearamide, a paraffin wax, an ester wax, a fatty alcohol wax, a long chain fatty alcohol, a fatty acid soap, a fatty acid ester, a silicon glycol, a fluorosilicone, a polyethylene glycol-polypropylene glycol copolymer, polydimethylsiloxane-silicon dioxide, ether, octyl alcohol, capryl alcohol, sorbitan trioleate, ethyl alcohol, 2-ethyl-hexanol, dimethicone, oleyl alcohol, simethicone, and combinations thereof.

[00502] In some embodiments, the pharmaceutical composition comprises a co-solvent. Illustrative examples of co-solvents include ethanol, poly(ethylene) glycol, butylene glycol, dimethylacetamide, glycerin, and propylene glycol.

[00503] In some embodiments, the pharmaceutical composition comprises a buffer. Illustrative examples of buffers include acetate, borate, carbonate, lactate, malate, phosphate, citrate, hydroxide, diethanolamine, monoethanolamine, glycine, methionine, guar gum, and monosodium glutamate.

[00504] In some embodiments, the pharmaceutical composition comprises a carrier or filler. Illustrative examples of carriers or fillers include lactose, maltodextrin, mannitol, sorbitol, chitosan, stearic acid, xanthan gum, and guar gum.

[00505] In some embodiments, the pharmaceutical composition comprises a surfactant. Illustrative examples of surfactants include *d*-alpha tocopherol, benzalkonium chloride, benzethonium chloride, cetrimide, cetylpyridinium chloride, docusate sodium, glyceryl behenate, glyceryl monooleate, lauric acid, macrogol 15 hydroxystearate, myristyl alcohol, phospholipids, polyoxyethylene alkyl ethers, polyoxyethylene sorbitan fatty acid esters, polyoxyethylene stearates, polyoxylglycerides, sodium lauryl sulfate, sorbitan esters, and vitamin E polyethylene(glycol) succinate.

[00506] In some embodiments, the pharmaceutical composition comprises an anti-caking agent. Illustrative examples of anti-caking agents include calcium phosphate (tribasic), hydroxymethyl cellulose, hydroxypropyl cellulose, and magnesium oxide.

[00507] Other excipients that may be used with the pharmaceutical compositions include, for example, albumin, antioxidants, antibacterial agents, antifungal agents, bioabsorbable polymers, chelating agents, controlled release agents, diluents, dispersing agents, dissolution enhancers, emulsifying agents, gelling agents, ointment bases, penetration enhancers, preservatives, solubilizing agents, solvents, stabilizing agents, and sugars. Specific examples of each of these agents are described, for example, in the *Handbook of Pharmaceutical Excipients*, Rowe et al. (Eds.) 6th Ed. (2009), The Pharmaceutical Press, incorporated by reference in its entirety.

[00508] In some embodiments, the pharmaceutical composition comprises a solvent. In some aspects, the solvent is saline solution, such as a sterile isotonic saline solution or dextrose solution. In some aspects, the solvent is water for injection.

[00509] In some embodiments, the pharmaceutical compositions are in a particulate form, such as a microparticle or a nanoparticle. Microparticles and nanoparticles may be formed from any suitable material, such as a polymer or a lipid. In some aspects, the microparticles or nanoparticles are micelles, liposomes, or polymersomes.

[00510] Further provided herein are anhydrous pharmaceutical compositions and dosage forms comprising an antibody conjugate, since, in some embodiments, water can facilitate the degradation of some antibodies.

[00511] Anhydrous pharmaceutical compositions and dosage forms provided herein can be prepared using anhydrous or low moisture containing ingredients and low moisture or low humidity conditions. Pharmaceutical compositions and dosage forms that comprise lactose and at least one active ingredient that comprises a primary or secondary amine can be anhydrous if substantial contact with moisture and/or humidity during manufacturing, packaging, and/or storage is expected.

[00512] An anhydrous pharmaceutical composition can be prepared and stored such that its anhydrous nature is maintained. Accordingly, anhydrous compositions can be packaged using materials known to prevent exposure to water such that they can be included in suitable formulary kits. Examples of suitable packaging include, but are not limited to, hermetically sealed foils, plastics, unit dose containers (*e.g.*, vials), blister packs, and strip packs.

[00513] Lactose-free compositions provided herein can comprise excipients that are well known in the art and are listed, for example, in the U.S. Pharmocopia (USP) SP (XXI)/NF (XVI). In general, lactose-free compositions comprise an active ingredient, a binder/filler, and a lubricant in pharmaceutically compatible and pharmaceutically acceptable amounts. Exemplary lactose-free dosage forms comprise an active ingredient, microcrystalline cellulose, pre gelatinized starch, and magnesium stearate.

[00514] Also provided are pharmaceutical compositions and dosage forms that comprise one or more excipients that reduce the rate by which an antibody or antibody-conjugate will decompose. Such excipients, which are referred to herein as "stabilizers," include, but are not limited to, antioxidants such as ascorbic acid, pH buffers, or salt buffers.

14.1. Parenteral Dosage Forms

[00515] In certain embodiments, provided are parenteral dosage forms. Parenteral dosage forms can be administered to subjects by various routes including, but not limited to, subcutaneous, intravenous (including bolus injection), intramuscular, and intraarterial. Because their administration typically bypasses subjects' natural defenses against contaminants, parenteral dosage forms are typically, sterile or capable of being sterilized prior to administration to a subject. Examples of parenteral dosage forms include, but are not limited to, solutions ready for injection, dry products ready to be dissolved or suspended in a pharmaceutically acceptable vehicle for injection, suspensions ready for injection, and emulsions.

[00516] Suitable vehicles that can be used to provide parenteral dosage forms are well known to those skilled in the art. Examples include, but are not limited to: Water for Injection USP; aqueous vehicles such as, but not limited to, Sodium Chloride Injection, Ringer's Injection, Dextrose Injection, Dextrose and Sodium Chloride Injection, and Lactated Ringer's Injection; water miscible vehicles such as, but not limited to, ethyl alcohol, polyethylene glycol, and polypropylene glycol; and non-aqueous vehicles such as, but not limited to, corn oil, cottonseed oil, peanut oil, sesame oil, ethyl oleate, isopropyl myristate, and benzyl benzoate.

[00517] Excipients that increase the solubility of one or more of the antibodies disclosed herein can also be incorporated into the parenteral dosage forms.

14.2. Dosage and Unit Dosage Forms

[00518] In human therapeutics, the doctor will determine the posology which he considers most appropriate according to a preventive or curative treatment and according to the age, weight, condition and other factors specific to the subject to be treated.

[00519] In certain embodiments, a composition provided herein is a pharmaceutical composition or a single unit dosage form. Pharmaceutical compositions and single unit dosage forms provided herein comprise a prophylactically or therapeutically effective amount of one or more prophylactic or therapeutic antibodies.

[00520] The amount of the antibody conjugate or composition which will be effective in the prevention or treatment of a disorder or one or more symptoms thereof will vary with the nature and severity of the disease or condition, and the route by which the antibody is administered. The frequency and dosage will also vary according to factors specific for each subject depending on the specific therapy (*e.g.*, therapeutic or prophylactic agents) administered, the severity of the disorder, disease, or condition, the route of administration, as well as age, body, weight, response, and the past medical history of the subject. Effective doses may be extrapolated from dose-response curves derived from *in vitro* or animal model test systems.

[00521] In certain embodiments, exemplary doses of a composition include milligram or microgram amounts of the antibody per kilogram of subject or sample weight (*e.g.*, about 10 micrograms per kilogram to about 50 milligrams per kilogram, about 100 micrograms per kilogram to about 25 milligrams per kilogram, or about 100 microgram per kilogram to about 10 milligrams per kilogram). In certain embodiment, the dosage of the antibody conjugate provided herein, based on weight of the antibody, administered to prevent, treat, manage, or ameliorate a disorder, or one or more symptoms thereof in a subject is 0.1 mg/kg, 1 mg/kg, 2 mg/kg, 3 m**g/kg, 4 mg/kg, 5 mg/kg, 6 mg/kg, 10 mg/kg, or 15 mg/kg or more of a subject's** body weight. In another embodiment, the dosage of the composition or a composition provided herein administered to prevent, treat, manage, or ameliorate a disorder, or one or more symptoms thereof in a subject is 0.1 mg to 200 mg, 0.1 mg to 100 mg, 0.1 mg to 50 mg, 0.1 mg to 25 mg, 0.1 mg to 20 mg, 0.1 mg to 15 mg, 0.1 mg to 10 mg, 0.1 mg to 7.5 mg, 0.1 mg to 5 mg, 0.1 to 2.5 mg, 0.25 mg to 20 mg, 0.25 to 15 mg, 0.25 to 12 mg, 0.25 to 10 mg, 0.25 mg to 7.5 mg, 0.25 mg to 5 mg, 0.25 mg to 2.5 mg, 0.5 mg to 20 mg, 0.5 to 15 mg, 0.5 to 12 mg, 0.5 to 10 mg, 0.5 mg to 7.5 mg, 0.5 mg to 5 mg, 0.5 mg to 2.5 mg, 1 mg to 20 mg, 1 mg to 15 mg, 1 mg to 12 mg, 1 mg to 10 mg, 1 mg to 7.5 mg, 1 mg to 5 mg, or 1 mg to 2.5 mg.

[00522] The dose can be administered according to a suitable schedule, for example, once, two times, three times, or for times weekly. It may be necessary to use dosages of the antibody conjugate outside the ranges disclosed herein in some cases, as will be apparent to those of ordinary skill in the art. Furthermore, it is noted that the clinician or treating physician will know how and when to interrupt, adjust, or terminate therapy in conjunction with subject response.

[00523] Different therapeutically effective amounts may be applicable for different diseases and conditions, as will be readily known by those of ordinary skill in the art. Similarly, amounts sufficient to prevent, manage, treat or ameliorate such disorders, but insufficient to cause, or sufficient to reduce, adverse effects associated with the antibodies provided herein are also encompassed by the herein described dosage amounts and dose frequency schedules. Further, when a subject is administered multiple dosages of a composition provided herein, not all of the dosages need be the same. For example, the dosage administered to the subject may be increased to improve the prophylactic or therapeutic effect of the composition or it may be decreased to reduce one or more side effects that a particular subject is experiencing.

[00524] In certain embodiments, treatment or prevention can be initiated with one or more loading doses of an antibody conjugate or composition provided herein followed by one or more maintenance doses.

[00525] In certain embodiments, a dose of an antibody conjugate or composition provided herein can be administered to achieve a steady-state concentration of the antibody in blood or serum of the subject. The steady-state concentration can be determined by measurement according to techniques available to those of skill or can be based on the physical characteristics of the subject such as height, weight and age.

[00526] In certain embodiments, administration of the same composition may be repeated and the administrations may be separated by at least 1 day, 2 days, 3 days, 5 days, 10 days, 15 days, 30 days, 45 days, 2 months, 75 days, 3 months, or 6 months. In other embodiments, administration of the same prophylactic or therapeutic agent may be repeated and the administration may be separated by at least 1 day, 2 days, 3 days, 5 days, 10 days, 15 days, 30 days, 45 days, 2 months, 75 days, 3 months, or 6 months.

14.3. Combination Therapies and Formulations

[00527] In certain embodiments, provided are compositions and therapeutic formulations comprising any of the antibody conjugates provided herein in combination with one or more

chemotherapeutic agents disclosed herein, and methods of treatment comprising administering such combinations to subjects in need thereof. Examples of chemotherapeutic agents include, but are not limited to, Erlotinib (TARCEVA®, Genentech/OSI Pharm.), Bortezomib (VELCADE®, Millennium Pharm.), Fulvestrant (FASLODEX®, AstraZeneca), Sutent (SU11248, Pfizer), Letrozole (FEMARA®, Novartis), Imatinib mesylate (GLEEVEC®, Novartis), PTK787/ZK 222584 (Novartis), Oxaliplatin (Eloxatin®, Sanofi), 5-FU (5-fluorouracil), Leucovorin, Rapamycin (Sirolimus, RAPAMUNE®, Wyeth), Lapatinib (TYKERB®, GSK572016, Glaxo Smith Kline), Lonafarnib (SCH 66336), Sorafenib (BAY43-9006, Bayer Labs), and Gefitinib (IRESSA®, AstraZeneca), AG1478, AG1571 (SU 5271; Sugen), alkylating agents such as thiotepa and CYTOXAN® cyclosphosphamide; alkyl sulfonates such as busulfan, improsulfan and piposulfan; aziridines such as benzodopa, carboquone, meturedopa, and uredopa; ethylenimines and methylamelamines including altretamine, triethylenemelamine, triethylenephosphoramide, triethylenethiophosphoramide and trimethylomelamine; acetogenins (especially bullatacin and bullatacinone); a camptothecin (including the synthetic analog topotecan); bryostatin; callystatin; CC-1065 (including its adozelesin, carzelesin and bizelesin synthetic analogs); cryptophycins (particularly cryptophycin 1 and cryptophycin 8); dolastatin; duocarmycin (including the synthetic analogs, KW-2189 and CB1-TM1); eleutherobin; pancratistatin; a sarcodictyin; spongistatin; nitrogen mustards such as chlorambucil, chlomaphazine, chlorophosphamide, estramustine, ifosfamide, mechlorethamine, mechlorethamine oxide hydrochloride, melphalan, novembichin, phenesterine, prednimustine, trofosfamide, uracil mustard; nitrosureas such as carmustine, chlorozotocin, fotemustine, lomustine, nimustine, and ranimnustine; antibiotics such as the enediyne antibiotics (*e.g.*, calicheamicin, especially uncialamycin, calicheamicin gammall, and calicheamicin omegall (Angew Chem. Intl. Ed. Engl. (1994) 33:183-186); dynemicin, including dynemicin A; bisphosphonates, such as clodronate; an esperamicin; as well as neocarzinostatin chromophore and related chromoprotein enediyne antibiotic chromophores), aclacinomysins, actinomycin, authramycin, azaserine, bleomycins, cactinomycin, carabicin, caminomycin, carzinophilin, chromomycinis, dactinomycin, daunorubicin, detorubicin, 6-diazo-5-oxo-L-norleucine, ADRIAMYCIN® (doxorubicin), morpholino-doxorubicin, cyanomorpholino-doxorubicin, 2-pyrrolino-doxorubicin and deoxydoxorubicin), epirubicin, esorubicin, idarubicin, marcellomycin, mitomycins such as mitomycin C, mycophenolic acid, nogalamycin, olivomycins, peplomycin, porfiromycin, puromycin, quelamycin, rodorubicin, streptonigrin, streptozocin, tubercidin, ubenimex, zinostatin, zorubicin; anti-metabolites such

as methotrexate and 5-fluorouracil (5-FU); folic acid analogs such as denopterin, methotrexate, pladienolide B, pteropterin, trimetrexate; purine analogs such as fludarabine, 6-mercaptopurine, thiamniprine, thioguanine; pyrimidine analogs such as ancitabine, azacitidine, 6-azauridine, carmofur, cytarabine, dideoxyuridine, doxifluridine, enocitabine, floxuridine; androgens such as calusterone, dromostanolone propionate, epitiostanol, mepitiostane, testolactone; anti-adrenals such as aminoglutethimide, mitotane, trilostane; folic acid replenisher such as frolinic acid; aceglatone; aldophosphamide glycoside; aminolevulinic acid; eniluracil; amsacrine; bestrabucil; bisantrene; edatraxate; defofamine; demecolcine; diaziquone; elformithine; elliptinium acetate; an epothilone; etoglucid; gallium nitrate; hydroxyurea; lentinan; lonidainine; maytansinoids such as maytansine and ansamitocins; mitoguazone; mitoxantrone; mopidanmol; nitraerine; pentostatin; phenamet; pirarubicin; losoxantrone; podophyllinic acid; 2-ethylhydrazide; procarbazine; PSK® polysaccharide complex (JHS Natural Products, Eugene, Oreg.); razoxane; rhizoxin; sizofuran; spirogermanium; tenuazonic acid; triaziquone; 2,2',2''-trichlorotriethylamine; trichothecenes (especially T-2 toxin, verracurin A, roridin A and anguidine); urethan; vindesine; dacarbazine; mannomustine; mitobronitol; mitolactol; pipobroman; gacytosine; arabinoside ("Ara-C"); cyclophosphamide; thiotepa; taxoids, *e.g.*, TAXOL® (paclitaxel; Bristol-Myers Squibb Oncology, Princeton, N.J.), ABRAXANE® (Cremophor-free), albumin-engineered nanoparticle formulations of paclitaxel (American Pharmaceutical Partners, Schaumberg, Ill.), and TAXOTERE® (doxetaxel; Rhone-Poulenc Rorer, Antony, France); chloranmbucil; GEMZAR® (gemcitabine); 6-thioguanine; mercaptopurine; methotrexate; platinum analogs such as cisplatin and carboplatin; vinblastine; etoposide (VP-16); ifosfamide; mitoxantrone; vincristine; NAVELBINE® (vinorelbine); novantrone; teniposide; edatrexate; daunomycin; aminopterin; capecitabine (XELODA®); ibandronate; CPT-11; topoisomerase inhibitor RFS 2000; difluoromethylornithine (DMFO); retinoids such as retinoic acid; and pharmaceutically acceptable salts, acids and derivatives of any of the above.

[00528] In certain embodiments, provided are compositions and therapeutic formulations comprising any of the antibody conjugates provided herein in combination with one or more PD-1 or PD-L1 inhibitors, and methods of treatment comprising administering such combinations to subjects in need thereof. In some embodiments, the one or more PD-1 or PD-L1 inhibitors comprise a small molecule blocker of the PD-1 or PD-L1 pathway. In some embodiments, the one or more PD-1 or PD-L1 inhibitors comprise an antibody that inhibits PD-1 or PD-L1 activity. In some embodiments, the one or more PD-1 or PD-L1 inhibitors are

selected from the group consisting of: CA-170, BMS-8, BMS-202, BMS-936558, CK-301, and AUNP12. In some embodiments, the one or more PD-1 or PD-L1 inhibitors are selected from the group consisting of: avelumab, nivolumab, pembrolizumab, atezolizumab, durvalumab, AMP-224 (GlaxoSmithKline), MEDI0680/AMP-514 (AstraZeneca), PDR001 (Novartis), cemiplimab, TSR-042 (Tesaro), Tizlelizumab/BGB-A317 (Beigene), CK-301 (Checkpoint Therapeutics), BMS-936559 (Bristol-Meyers Squibb), camrelizumab, sintilimab, toripalimab, genolimzumab, and A167 (Sichuan Kelun-Biotech Biopharmaceutical). In some embodiments, the one or more PD-1 or PD-L1 inhibitors are selected from the group consisting of: MGA012 (Incyte/MacroGenics), PF-06801591 (Pfizer/Merck KGaA), LY3300054 (Eli Lilly), FAZ053 (Novartis), PD-11 (Novartis), CX-072 (CytomX), BGB-A333 (Beigene), BI 754091 (Boehringer Ingelheim), JNJ-63723283 (Johnson and Johnson/Jannsen), AGEN2034 (Agenus), CA-327 (Curis), CX-188 (CytomX), STI –A1110 (Servier), JTX-4014 (Jounce), (LLY) AM0001 (Armo Biosciences), CBT-502 (CBT Pharmaceuticals), FS118 (F-Star/Merck KGaA), XmAb20717 (Xencor), XmAb23104 (Xencor), AB122 (Arcus Biosciences), KY1003 (Kymab), RXI-762 (RXi). In some embodiments, the one or more PD-1 or PD-L1 inhibitors are selected from the group consisting of: PRS-332 (Pieris Pharmaceuticals), ALPN-202 (Alpine Immune Science), TSR-075 (Tesaro/Anaptys Bio), MCLA-145 (Merus), MGD013 (Macrogenics), MGD019 (Macrogenics). In some embodiments, the one or more PD-1 or PD-L1 inhibitors are selected from an anti-PD1 mono-specific or bi-specific antibody described in, for example, WO 2016/077397, WO 2018/156777, and International Application No. PCT/US2013/034213, filed May 23, 2018.

[00529] The agents administered in combination with the antibody conjugates disclosed herein can be administered just prior to, concurrent with, or shortly after the administration of the antibody conjugates. For purposes of the present disclosure, such administration regimens are considered the administration of an **antibody conjugate "in combination with" an additional** therapeutically active component. Embodiments include pharmaceutical compositions in which an antibody conjugate disclosed herein is co-formulated with one or more of the chemotherapeutic agents, PD-1 inhibitors, or PD-L1 inhibitors disclosed herein.

15. Therapeutic Applications

[00530] For therapeutic applications, the antibody conjugates provided herein can be administered to a mammal, generally a human, in a pharmaceutically acceptable dosage form such as those known in the art and those discussed above. For example, the antibody conjugates

may be administered to a human intravenously as a bolus or by continuous infusion over a period of time, by intramuscular, intraperitoneal, intra-cerebrospinal, subcutaneous, intra-articular, intrasynovial, intrathecal, or intratumoral routes. The antibody conjugates also are suitably administered by peritumoral, intralesional, or perilesional routes, to exert local as well as systemic therapeutic effects. The intraperitoneal route may be particularly useful, for example, in the treatment of ovarian tumors.

[00531] The antibody conjugates provided herein may be useful for the treatment of any disease or condition involving folate receptor alpha (FOLR1). In some embodiments, the disease or condition is a disease or condition that can be diagnosed by overexpression of folate receptor alpha. In some embodiments, the disease or condition is a disease or condition that can benefit from treatment with an anti-folate receptor alpha antibody. In some embodiments, the disease or condition is a cancer.

[00532] Any suitable cancer may be treated with the antibody conjugates provided herein. Illustrative suitable cancers include, for example, acute lymphoblastic leukemia (ALL), acute myeloid leukemia (AML), adrenocortical carcinoma, anal cancer, appendix cancer, astrocytoma, basal cell carcinoma, brain tumor, bile duct cancer, bladder cancer, bone cancer, breast cancer (including triple-negative breast cancer, or TNBC), bronchial tumor, carcinoma of unknown primary origin, cardiac tumor, cervical cancer, chordoma, colon cancer, colorectal cancer, craniopharyngioma, ductal carcinoma, embryonal tumor, endometrial cancer, ependymoma, esophageal cancer, esthesioneuroblastoma, fallopian tube carcinoma, fibrous histiocytoma, Ewing sarcoma, eye cancer, germ cell tumor, gallbladder cancer, gastric cancer, gastrointestinal carcinoid tumor, gastrointestinal stromal tumor, gestational trophoblastic disease, glioma, head and neck cancer, hepatocellular cancer, histiocytosis, Hodgkin lymphoma, hypopharyngeal cancer, intraocular melanoma, islet cell tumor, Kaposi sarcoma, kidney cancer, Langerhans cell histiocytosis, laryngeal cancer, lip and oral cavity cancer, liver cancer, lobular carcinoma *in situ*, lung cancer, macroglobulinemia, malignant fibrous histiocytoma, melanoma, Merkel cell carcinoma, mesothelioma, metastatic squamous neck cancer with occult primary, midline tract carcinoma involving *NUT* gene, mouth cancer, multiple endocrine neoplasia syndrome, multiple myeloma, mycosis fungoides, myelodysplastic syndrome, myelodysplastic/myeloproliferative neoplasm, nasal cavity and par nasal sinus cancer, nasopharyngeal cancer, neuroblastoma, non-small cell lung cancer (NSCLC), oropharyngeal cancer, osteosarcoma, ovarian cancer, pancreatic cancer, papillomatosis, paraganglioma, parathyroid cancer, penile cancer, pharyngeal cancer,

pheochromocytomas, pituitary tumor, pleuropulmonary blastoma, primary central nervous system lymphoma, primary peritoneal carcinoma, prostate cancer, rectal cancer, renal cell cancer, renal pelvis and ureter cancer, retinoblastoma, rhabdoid tumor, salivary gland cancer, Sezary syndrome, skin cancer, small cell lung cancer, small intestine cancer, soft tissue sarcoma, spinal cord tumor, stomach cancer, T-cell lymphoma, teratoid tumor, testicular cancer, throat cancer, thymoma and thymic carcinoma, thyroid cancer, urethral cancer, uterine cancer, vaginal cancer, vulvar cancer, and Wilms tumor.

[00533] In some embodiments, the disease to be treated with the antibody conjugates provided herein is gastric cancer, colorectal cancer, renal cell carcinoma, cervical cancer, non-small cell lung carcinoma, ovarian cancer, uterine cancer, fallopian tube carcinoma, primary peritoneal carcinoma, uterine corpus carcinoma, endometrial carcinoma, prostate cancer, breast cancer, head and neck cancer, brain carcinoma, liver cancer, pancreatic cancer, mesothelioma, and/or a cancer of epithelial origin. In particular embodiments, the disease is colorectal cancer. In some embodiments, the disease is ovarian cancer. In some embodiments, the disease is breast cancer. In some embodiments, the disease is triple-negative breast cancer (TNBC). In some embodiments, the disease is lung cancer. In some embodiments, the disease is non-small cell lung cancer (NSCLC). In some embodiments, the disease is head and neck cancer. In some embodiments, the disease is renal cell carcinoma. In some embodiments, the disease is brain carcinoma. In some embodiments, the disease is endometrial cancer.

16. Diagnostic Applications

[00534] In some embodiments, the antibody conjugates provided herein are used in diagnostic applications. For example, an anti-FOLR1 antibody conjugate may be useful in assays for FOLR1 protein. In some aspects the antibody conjugate can be used to detect the expression of FOLR1 in various cells and tissues. These assays may be useful, for example, in making a diagnosis and/or prognosis for a disease, such as a cancer.

[00535] In some diagnostic and prognostic applications, the antibody conjugate may be labeled with a detectable moiety. Suitable detectable moieties include, but are not limited to radioisotopes, fluorescent labels, and enzyme-substrate labels. In another embodiment, the anti-FOLR1 antibody conjugate need not be labeled, and the presence of the antibody conjugate can be detected using a labeled antibody which specifically binds to the anti-FOLR1 antibody conjugate.

17. Affinity Purification Reagents

[00536] The antibody conjugates provided herein may be used as affinity purification agents. In this process, the antibody conjugates may be immobilized on a solid phase such a resin or filter paper, using methods well known in the art. The immobilized antibody conjugate is contacted with a sample containing the folate receptor alpha protein (or fragment thereof) to be purified, and thereafter the support is washed with a suitable solvent that will remove substantially all the material in the sample except the folate receptor alpha protein, which is bound to the immobilized antibody. Finally, the support is washed with another suitable solvent, such as glycine buffer, pH 5.0 that will release the folate receptor alpha protein from the antibody.

18. Kits

[00537] In some embodiments, an anti-FOLR1 antibody conjugate provided herein is provided in the form of a kit, i.e., a packaged combination of reagents in predetermined amounts with instructions for performing a procedure. In some embodiments, the procedure is a diagnostic assay. In other embodiments, the procedure is a therapeutic procedure.

[00538] In some embodiments, the kit further comprises a solvent for the reconstitution of the anti-FOLR1 antibody conjugate. In some embodiments, the anti-FOLR1 antibody conjugate is provided in the form of a pharmaceutical composition.

EXAMPLES

EXAMPLE 1

GENERATION AND PRIMARY SCREENING OF ANTI-FOLR1 ANTIBODIES

[00539] Antibody Fab libraries were constructed using a standard overlap extension PCR protocol with mutagenic primers targeting complementary determining regions (CDRs). *See* Heckman and Pease, *Nat. Protoc.*, 2007, 2:924-932; Stafford *et al.*, 2014, *Protein Eng. Des. Sel.* 27:97-109, both incorporated by reference in their entireties. Selections for novel antibodies were performed using standard ribosome display protocols. *See* Dreier and Plückthun, 2011, *Methods Mol Biol* 687:283-306, which is incorporated herein by reference in its entirety.

[00540] Initial antibody leads from ribosome display were derived from a naïve human library which was constructed by overlapping PCR using trastuzumab HC as the base template. CDRs H1 and H2 were randomized with the same design as described by Lee *et al.*, *J. Mol.*

Biol. 2004, 340:1073-1093 using oligonucleotides purchased from Integrated DNA Technologies. In this design, CDRs H1 and H2 closely match the observed amino acid distributions of natural human antibodies. CDR H3 was diversified using oligonucleotides incorporating trimer phosphoramidite mixtures (TRIMs) for amino acid randomization. The TRIM oligos were synthesized as described by Yagodkin A *et al., Nucleosides Nucleotides Nucleic Acids* 2007, 26:473-97. Specifically, six separate oligonucleotides containing TRIMs were used to make 6 separate H3 loop-lengths (13-18; as defined by Zemlin *et al.*) to match the most common loop lengths observed in the human repertoire. Together these loop lengths comprise approximately 54.5% of the naturally-occurring loop length variation in human IgGs as reported by Zemlin *et al.*, *J. Mol. Biol.* 2003, 334:733-749. The frequency distribution of each amino acid was designed to closely match the observed distribution of amino acids in CDR H3 of human IgGs as reported by Zemlin *et al.* Altogether, the library closely matches natural human antibody variation which is known in the field to improve antibody stability and folding of antibodies as described by Zhai *et al., J Mol Biol.* 2011, 412:55-71. The heavy chain (HC) library was paired with a constant, unmodified trastuzumab light chain (LC) throughout the selection process as described by Stafford *et al.*, *Protein Eng Des Sel* 2014, 27:97-109.

[00541] Affinity maturated antibody leads (*e.g.*, SRP 1848 antibodies, below) were derived from a focused library, biased towards two leads, which was constructed by overlapping PCR **using "soft-randomized" oligonucleotides purchased from Eurofins MWG Operon. Soft-**randomization is a process in which a biased distribution of nucleotides is used for each soft-randomized codon such that the parent amino acid sequence is coded more frequently than other amino acids ~30% of the time. Other amino acids are coded at each position but at a lower percentage. At each soft-randomized position, 70% of the parent nucleotide is mixed with 10% of the other three nucleotides. For the library, CDRs H1, H2, and H3 were soft-randomized simultaneously and selected by standard ribosome display protocols. As with the selection of initial leads, the affinity matured antibodies were paired with a constant, unmodified trastuzumab LC throughout the selection process as described by Stafford *et al.*, *Protein Eng Des Sel* 2014, 27:97-109.

[00542] Selections for novel antibodies were performed using standard ribosome display protocols. *See* Dreier and Plückthun, *Methods Mol. Biol.*, 2003, 687:283-306, Clifton, NJ, incorporated by reference in its entirety. Fab ribosome display selections were performed according to published protocols. *See* Stafford *et al.*, 2014, *Protein Eng. Des. Sel.* 27:97-109; Hanes and Plückthun, *Proc. Natl. Acad. Sci. U.S.A.*, 1997, 94:4937-4942; both incorporated by

reference in their entireties. After multiple rounds of selection, the DNA from RT-PCR output was cloned into an optimized vector for cell-free expression using standard molecular biology techniques. *See* Yin et al., *mAbs*, 2012, 4:217-225, incorporated by reference in its entirety. All constructs were HIS- and FLAG-tagged to streamline purification and testing during screening.

[00543] Libraries of antibody variants generated by selection workflow were transformed into *E. coli* and grown on agar plates with antibiotic (kanamycin). Individual colonies were grown in liquid broth (TB + kanamycin), and used as a template for DNA amplification via rolling circle amplification (RCA). The variants were then expressed in cell-free protein synthesis reactions as described in Yin et al., *mAbs*, 2012, 4:217-225.

[00544] Briefly, cell-free extracts were treated with 50 μM iodoacetamide for 30 min at room temperature (20°C) and added to a premix containing cell-free components (*see* Cai *et al.*, *Biotechnol Prg*, 2015, 3:823-831, incorporated by reference in its entirety) and 10% (v/v) **RCA DNA template (approximately 10 μg/mL DNA) for HC variants, in addition to 2.5 ug/mL** Trastuzumab LC which is present for antibody assembly but is not varied in the library. Sixty microliters of cell-free reactions were incubated at 30°C for 12 hr on a shaker at 650 rpm in 96-well plates. Four hundred to one-thousand-five-hundred (400 to 1500) colonies were screened, depending on the predicted diversity of different selection campaigns.

[00545] Following synthesis, each reaction was diluted 1:50 into PBS (pH 7.4) with 3% fetal bovine serum (FBS), and expressed variants were tested for functional activity via cell-based ELISA binding to CHO-hFOLR1 cells (human FOLR1 expressed recombinantly in Chinese Hamster Ovary cells). Briefly, 384-well plates were seeded with CHO-control or CHO-hFOLR1 cells the day before the assay. On the day of the assay, cells were fixed with 20 uL of 4% paraformaldehyde in PBS for 15 minutes in the dark, washed with PBS, and then blocked with 30% FBS in PBS for 30 minutes at room temperature. Antibody variants of interest (1:50 diluted cell-free reaction) were allowed to bind to the fixed CHO-hFOLR1 cells, and detected with secondary antibodies (e.g. HRP-conjugated Anti-human Fc or anti-FLAG) and then detected with chemiluminescent substrate (Pierce ELISA SuperSignal™ Substrate). Chemiluminescence was quantified on a Molecular Devices SpectraMax® M5 plate reader. Top hits were selected based on cell-based ELISA signal/noise ratio, and their nucleotides were sequenced. Based on binding activity and sequence analysis, a subset of variants was selected for further scale-up and characterization.

[00546] The top leads from ELISA-based screening were cultured, and plasmid minipreps were performed using a QIAprep® 96 Turbo miniprep kit (Qiagen) according to the manufacturer's instructions. 10 μg/mL miniprepped DNA was added to 4 mL cell-free reactions and incubated overnight for 12 hr at 30°C, at 650 rpm. In the case of IgG variants with a common Trastuzumab LC, 7.5 ug/mL of the HC variant DNA and 2.5 ug/mL of the common Trastuzumab LC were added to the reaction.

[00547] Expressed variants from clarified cell-free reactions were purified via immobilized metal ion affinity chromatography (IMAC) purification using a semi-automated high throughput batch purification method. Briefly, purifications were performed in a 96-well plate format where 50 μL/well of IMAC resin (Ni Sepharose High Performance, GE Healthcare) was equilibrated in IMAC binding buffer (50 mM Tris pH 8.0, 300 mM NaCl, 10 mM imidazole), incubated with 1 mL cell-free reaction for 15 minutes followed by two washes in IMAC binding buffer. His-tagged antibody variants were then eluted using 200 μL IMAC elution buffer (50 mM Tris pH 8.0, 300 mM NaCl, 500 mM imidazole) and buffer exchanged into PBS using a 96-well Zeba plate (7 kD MWCO, Thermo Fisher). Purified antibodies were quantified via high throughput capillary electrophoresis using the LabChip GXII (Perkin Elmer) against a Herceptin standard curve, according to the manufacturer's instructions.

[00548] Exemplary affinity-matured antibodies are reported in Table 5, below.

Table 5. Affinity Matured (SRP1848) Antibodies

Antibody	VH	SEQ ID NO.	VL	SEQ ID NO.
1	SRP1848-A01	308	Trastuzumab	367
2	SRP1848-A02	309	Trastuzumab	367
3	SRP1848-A04	310	Trastuzumab	367
4	SRP1848-A06	311	Trastuzumab	367
5	SRP1848-A07	312	Trastuzumab	367
6	SRP1848-A08	313	Trastuzumab	367
7	SRP1848-A09	314	Trastuzumab	367
8	SRP1848-A10	315	Trastuzumab	367
9	SRP1848-B01	316	Trastuzumab	367
10	SRP1848-B03	317	Trastuzumab	367
11	SRP1848-B04	318	Trastuzumab	367
12	SRP1848-B05	319	Trastuzumab	367
13	SRP1848-B06	320	Trastuzumab	367
14	SRP1848-B07	321	Trastuzumab	367

Antibody	VH	SEQ ID NO.	VL	SEQ ID NO.
15	SRP1848-B09	322	Trastuzumab	367
16	SRP1848-B10	323	Trastuzumab	367
17	SRP1848-B11	324	Trastuzumab	367
18	SRP1848-C01	325	Trastuzumab	367
19	SRP1848-C03	326	Trastuzumab	367
20	SRP1848-C04	327	Trastuzumab	367
21	SRP1848-C05	328	Trastuzumab	367
22	SRP1848-C07	329	Trastuzumab	367
23	SRP1848-C10	330	Trastuzumab	367
24	SRP1848-D02	331	Trastuzumab	367
25	SRP1848-D03	332	Trastuzumab	367
26	SRP1848-D04	333	Trastuzumab	367
27	SRP1848-D05	334	Trastuzumab	367
28	SRP1848-D07	335	Trastuzumab	367
29	SRP1848-D09	336	Trastuzumab	367
30	SRP1848-D10	337	Trastuzumab	367
31	SRP1848-E01	338	Trastuzumab	367
32	SRP1848-E02	339	Trastuzumab	367
33	SRP1848-E03	340	Trastuzumab	367
34	SRP1848-E05	341	Trastuzumab	367
35	SRP1848-E06	342	Trastuzumab	367
36	SRP1848-E07	343	Trastuzumab	367
37	SRP1848-F01	344	Trastuzumab	367
38	SRP1848-F02	345	Trastuzumab	367
39	SRP1848-F04	346	Trastuzumab	367
40	SRP1848-F05	347	Trastuzumab	367
41	SRP1848-F06	348	Trastuzumab	367
42	SRP1848-F07	349	Trastuzumab	367
43	SRP1848-F08	350	Trastuzumab	367
44	SRP1848-F09	351	Trastuzumab	367
45	SRP1848-F10	352	Trastuzumab	367
46	SRP1848-F11	353	Trastuzumab	367
47	SRP1848-G01	354	Trastuzumab	367
48	SRP1848-G03	355	Trastuzumab	367
49	SRP1848-G04	356	Trastuzumab	367
50	SRP1848-G06	357	Trastuzumab	367
51	SRP1848-G07	358	Trastuzumab	367
52	SRP1848-G09	359	Trastuzumab	367
53	SRP1848-G10	360	Trastuzumab	367
54	SRP1848-G11	361	Trastuzumab	367

Antibody	VH	SEQ ID NO.	VL	SEQ ID NO.
55	SRP1848-H01	362	Trastuzumab	367

EXAMPLE 2

PREPARATION OF SCFVS

[00549] A single-chain antibody is made in either the V_HV_L or V_LV_H orientation with a linker sequence between the V_H and V_L domains. Typically scFv linkers are composed of (GGGGS)n repeats where n = 3, 4, 5, or 6 for linkers of 15, 20, 25, or 30 residues respectively. For cell-free expression, an N-terminal Met is added, but for mammalian expression a leader peptide is added. On the C-terminal end of the scFv, an Fc sequence can be added to extend *in vivo* half-life or the scFv can be used directly. An optional linker sequence can be incorporated between the scFv and the Fc. An exemplary scFv-Fc linker sequence is AAGSDQEPKSS (SEQ ID NO: 378). C-terminal affinity tags can optionally be added to facilitate purification and assay development. An exemplary affinity tag is a C-terminal FlagHis tag GSGDYKDDDDKGSGHHHHHH (SEQ ID NO: 376). A stop codon is typically inserted at the end of the sequence. An exemplary scFv can include an N-terminal Met residue, a V_H domain, a GGGGSGGGGSGGGGS (SEQ ID NO: 377) linker, a V_L domain, an AAGSDQEPKSS (SEQ ID NO: 378) linker, an Fc domain, a FlagHis tag, and a stop codon.

EXAMPLE 3

AFFINITY AND KINETIC BINDING ANALYSES

[00550] Anti-Fc polyclonal antibodies were immobilized onto a CM5 chip (GE Life Sciences) using amine coupling chemistry (from Amine Coupling Kit, GE Life Sciences). The immobilization steps were carried out at a flow rate of 25 µL/min in 1x HBS-EP+ buffer (GE Life Sciences; 10x Stock diluted before use). The sensor surfaces were activated for 7 min with a mixture of N-hydroxysuccinimide (NHS, 0.05 M) and 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide (EDC, 0.2 M). The anti-Fc polyclonal antibodies were injected over all 4 flow cells at a concentration of 25 µg/mL in 10 mM sodium acetate, pH 4.5, for 7 min. Ethanolamine (1 M, pH 8.5) was injected for 7 min to block any remaining activated groups. An average of 12,000 response units (RU) of capture antibody was immobilized on each flow cell.

[00551] Off-rate and kinetic binding experiments were performed at 25°C using 1x HBS-EP+ buffer. Test and control antibodies were injected over the anti-Fc surface at concentrations of 5-10 µg/mL for 12 seconds at a flow rate of 10 µL/min on flow cells 2, 3 and 4, followed by

a buffer wash for 30 seconds at the same flow rate. Kinetic characterization of antibody samples was carried out with a single concentration of antigen (for off-rate ranking) or a 1:2 dilution series of antigen (for kinetic characterization) and 1 injection of 0 nM antigen. After capturing ligand (antibody) on the anti-Fc surface, the analyte (human FOLR1-HIS) was bound at 50, 25, 12.5, 6.25 and 0 nM for 180 seconds, followed by a 600 second dissociation phase at a flow rate of 50 µL/min. Between each ligand capture and analyte binding cycle, regeneration was carried out using 2 injections of 10 mM glycine pH 2.0 for 30 seconds at 30 µL/min, followed by a 30 second buffer wash step.

[00552] The data were fit with the Biacore T200 Evaluation software, using a 1:1 Langmuir binding model. K_D (affinity, nM) was determined as a ratio of the kinetic rate constants calculated from the fits of the association and dissociation phases.

EXAMPLE 4

FLOW CYTOMETRY-BASED CELL BINDING ASSAY

[00553] Variants with expression levels >250 nM were tested in a fluorescence-activated cell sorting (FACS) cell-binding assay. CHO cells were transfected to stably express human (CHO-hFOLR1), cynomolgus (CHO-cFOLR1), or mouse (CHO-mFOLR1) target molecule FOLR1 on the cell surface. Parental CHO cells were used as a negative control to determine background binding levels. Parental CHO and stably transfected CHO-hFOLR1, CHO-cFOLR1, and CHO-mFOLR1 cells were cultured in Ham's F-12: high glucose DMEM (50:50) (Corning, Cellgro-Mediatech) supplemented with 10% heat-inactivated fetal bovine serum (Corning, Cellgro-Mediatech), 1% Penicillin/Streptomycin (Corning, Cellgro-Mediatech) and 2 mmol/L-glutamax (Life Technology).

[00554] A mixture of fluoresecent-labeled parental CHO cells and unlabeled CHO-hFOLR1cells were prepared as follows. Parental CHO cells were washed twice in PBS and incubated in PBS containing with 1 nM CellTrace™ Oregon Green488® (Life Technologies) at 37°C for 30 minutes. Labeled parental CHO cells were then washed 2x with Ham's F-12 media and 2x with FACS buffer (PBS with 1% bovine serum albumin). Unlabeled CHO-hFOLR1 cells were similarly washed and prepared. Labeled parental CHO and unlabeled CHO-hFOLR1 cells were combined at 1:1 ratio and seeded at 50 µL per well (200,000 cells per well) in 96 well polypropylene plates. Cells were mixed with 50 µL of test antibodies (*i.e.*, anti-FOLR1 variants) serially diluted in FACS buffer and incubated on ice for 60 mins. Cells were washed with FACS buffer and incubated on ice for 60 mins with 100 µL FACS buffer

containing 2.5 μg/mL R-Phycoerythrin-conjugated goat anti-Human IgG (Jackson ImmunoResearch Laboratories, West Grove, PA). Cells were washed twice with FACS buffer, fixed in 2% paraformaldehyde in PBS (Santa Cruz Biotechnology; Dallas, TX) for 10 mins on ice in the dark, and analyzed using the BD LSR II Flow Cytometer (BD Biosciences; San Jose, CA). Data were analyzed using FlowJo® software (FlowJo, LLC; Ashland, OR) to determine mean fluorescence intensities. Binding constants were calculated using the statistical software, GraphPad Prism (GraphPad Software; La Jolla, CA) using the nonlinear regression equation, one site – specific binding with Hill slope. Secondary antibody alone was used as a control, in addition to measuring non-specific antibody binding to CHO parental cells.

[00555] This procedure was repeated to assess cell binding in CHO-cFOLR1 and CHO-mFOLR1 cells.

EXAMPLE 5

CELL-KILLING ANALYSIS

[00556] The internalization of the antibodies was evaluated by a secondary antibody cell killing assay on target positive cells. FOLR1-positive KB cells were obtained from ATCC, and FOLR1-positive Igrov1 cells were obtained from NIH. The cells were maintained in Ham's F-12: high glucose DMEM (50:50) (Corning, Cellgro-Mediatech) supplemented with 10% heat-inactivated fetal bovine serum (Corning, Cellgro-Mediatech, Manassas, Virginia), 1% Penicillin/Streptomycin (Corning, Cellgro-Mediatech, Manassas, Virginia) and 2 mmol/L-glutamax (Thermo Fisher Scientific, Waltham, Massachusetts). Adherent cells were washed twice with calcium and magnesium-free Hanks Balanced Salt Solution (HBSS), harvested with HYQ®TASE™ (Hyclone; Thermo Fisher Scientific, Waltham, Massachusetts) and counted by the Vi-CELL Cell Viability Analyzers (Beckman Coulter, Indianapolis, Indiana). A total of 625 cells were seeded in each well of a 384-well flat bottom white polystyrene plate. Lead antibodies were formulated at 4-fold starting concentration in the cell culture medium and filtered through MultiScreenHTS 96-Well Filter Plates (Millipore; Billerica, Massachusetts). Serial dilutions of test antibody (1:3 serial dilution starting from 200 nM) was added into treatment wells, and an anti-human Fc nanobody conjugated to hemiasterlin via a cleavable linker was then added into each well at a fixed final concentration of 20 nM. Assay plates were cultured at 37°C in a CO_2 incubator for 120 hrs before assay. For cell viability measurement, 30 μL of Cell Titer-Glo® reagent (Promega Corp. Madison, WI) was added into each well, and plates were processed as per product instructions. Relative luminescence was measured on an

ENVISION® plate reader (Perkin-Elmer; Waltham, MA). Relative luminescence readings were converted to percent viability using untreated cells as controls. Data was fitted with non-linear regression analysis, using a log(inhibitor) vs. response-variable slope, 4 parameter fit with GraphPad Prism (GraphPad v 5.0, Software; San Diego, California). Data was expressed as relative cell viability (ATP content) % vs. dose of antibody.

EXAMPLE 6

GENERATION OF HYBRIDOMA

[00557] Immunocompetent mice (C57BL/6) were immunized with mouse MC38 cells overexpressing human FOLR1. FOLR1-specific antibodies were detected in the sera, and the spleen was harvested and fused with P3X cells to generate the hybridomas (Aragen Biosciences, Morgan Hill, CA), similar to what has been previously described. *See* Chronopoulou, *et al.*, 2014, *Methods Mol Biol* 1131:47-70, and Kim, *et al.*, 2014, *Methods Mol Biol* 1131:31-45, each of which is incorporated herein by reference in its entirety. Total RNA was extracted from hybridoma cells using QIAGEN RNeasy Mini Kit (Cat No. 74104) and converted to cDNA using a Clontech SMARTer RACE cDNA Amplification Kit (Cat. No. 634923) (Lake Pharma, Belmont, CA). Positive clones were identified by gel electrophoresis, cloned using an Invitrogen TOPO kit, and sequenced using standard Sanger methods. The CDRs for m6D1 were grafted onto human antibody frameworks VH1-18, VH3-33, VH2-5, VH2-70, VH4-30-4, Vk1-5, Vk3-11, Vk2-30, Vk1-33, and Vk1-16 by standard methodology to yield humanized antibodies. *See* Kuramochi, *et al.*, 2014, *Methods Mol Biol* 1060:123-137, which is incorporated herein by reference in its entirety. Of these grafts, the h6D1-HC3/LC4 (VH3-33/Vk3-11 grafts) and h6D1-HC3/LC5 (VH3-33/Vk1-5 grafts) IgGs gave the best yield when expressed in cell-free and maintained the highest affinity. Both HC3/LC4 and HC3/LC5 humanized variants were progressed into affinity maturation by Fab-based ribosome display (as described above) targeting the heavy chain CDRs by soft-randomization leaving the light-chain constant, as described in Stafford, *et al.*, 2014, *Protein Eng Des Sel* 4:97-109, which is incorporated herein by reference in its entirety.

[00558] Certain antibodies were generated by affinity maturation of humanized mouse antibodies. Exemplary antibody candidates are reported in Table 6, below.

Table 6. Affinity-matured humanized antibodies (SRP2060).

Antibody	VH	SEQ ID NO.	VL	SEQ ID NO.
56	SRP2060-E10	363	H6D1-LC4	368
57	SRP2060-E05	364	H6D1-LC4	368
58	SRP2060-B01	365	H6D1-LC5	369
59	SRP2060-A06	366	H6D1-LC5	369

EXAMPLE 7

CHARACTERISTICS OF ILLUSTRATIVE ANTI-FOLR1 ANTIBODIES

[00559] Tables 7 through 9 show results obtained using the illustrative antibodies described herein.

[00560] Table 7 shows results obtained with antibodies isolated from affinity-maturation of initial antibody leads obtained from a naïve Fab TRiM ribosome display library, constructed on a Trastuzumab heavy chain (HC) framework.

[00561] Table 8 shows kinetic binding results obtained for the same antibodies listed in Table 7.

[00562] Table 9 shows results obtained from antibodies isolated from humanized mouse clone candidates.

Table 7. Affinity-matured antibodies from initial leads (Trastuzumab HC framework).

Fab-HC Variant ID	KB, 2° Antibody Cell Killing, Nb-239		Igrov, 2° Antibody Cell Killing, Nb-SC239		CHO-human FolR1		CHO-cyno FolR1		CHO-mouse FolR1	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	Bmax (MFI)	K_D (nM)	Bmax (MFI)	K_D (nM)	Bmax (MFI)	K_D (nM)
SRP1848-A01	0.064	94	0.015	71	25899	0.39	22552	0.44	16475	0.8
SRP1848-A02	0.028	94	0.039	71	24710	0.64	18500	0.50	18569	2.4
SRP1848-A07	0.062	95	0.029	68	29182	0.61	23643	0.43	9646	0.9
SRP1848-C03	0.074	93	0.035	72	29143	0.51	25148	0.50	3310	3.0
SRP1848-F04	0.096	93	0.015	73	26867	0.73	26353	0.55	2741	11.0
SRP1848-B04	0.035	94	0.018	70	27818	0.72	27796	0.65	2187	17.9
SRP1848-B11	0.058	93	0.026	74	28394	0.56	22885	0.34	1632	3.9
SRP1848-F07	0.057	92	0.018	71	27371	0.58	18662	0.56	1387	8.8
SRP1848-E06	0.060	93	0.025	74	25611	0.48	15755	0.26	2349	1.2
SRP1848-A09	0.060	93	0.026	71	28910	0.61	20248	0.31	7990	1.0
SRP1848-E07	0.059	94	0.013	73	27284	0.54	20381	0.23	11837	1.2
SRP1848-G03	0.064	91	0.021	76	26424	0.82	19238	0.44	2220	2.4
SRP1848-A04	0.052	92	0.015	64	26810	0.43	23055	0.30	3888	2.0
SRP1848-H01	0.049	96	0.016	67	26985	0.59	17227	0.28	3950	33.8
SRP1848-B10	0.040	97	0.020	71	28186	0.83	21268	0.44	2455	7.3
SRP1848-C07	0.065	93	0.013	67	28757	0.62	18136	0.23	3170	1.4
SRP1848-F05	0.061	94	0.015	74	27155	0.72	24731	0.61	5100	18.0
SRP1848-D02	0.034	93	0.027	71	28804	0.60	27973	0.61	916	87.0
SRP1848-A08	0.039	93	0.013	65	28554	0.62	26197	0.45	3202	2.5
SRP1848-E03	0.057	94	0.027	73	26694	0.76	17427	0.43	5939	0.5
SRP1848-A10	0.033	96	0.027	75	27097	0.66	14816	0.47	10167	1.2

Fab-HC Variant ID	KB, 2° Antibody Cell Killing, Nb-239		Igrov, 2° Antibody Cell Killing, Nb-SC239		CHO-human FolR1		CHO-cyno FolR1		CHO-mouse FolR1	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	Bmax (MFI)	KD (nM)	Bmax (MFI)	KD (nM)	Bmax (MFI)	KD (nM)
SRP1848-F10	0.038	94	0.009	68	25554	0.36	20700	0.40	1742	6.9
SRP1848-D05	0.055	92	0.030	73	26748	0.57	22202	0.45	1360	14.0
SRP1848-C01	0.060	90	0.023	68	28527	0.66	25941	0.60	1369	26.0
SRP1848-F01	0.047	91	0.018	69	25240	0.56	21491	0.43	3750	1.8
SRP1848-D04	0.380	97	0.068	77	29297	2.21	25737	2.84	NB	NB
SRP1848-E05	0.071	95	0.027	78	27306	0.46	28170	0.55	NB	NB
SRP1848-A06	0.046	93	0.020	72	24521	0.47	20170	0.30	2767	2.4
SRP1848-B01	0.064	95	0.031	82	26634	1.06	23881	0.83	3404	16.4
SRP1848-C04	0.006	94	0.016	68	26269	0.44	22014	0.86	2506	62.0
SRP1848-C10	0.057	96	0.036	75	27465	0.91	15966	0.27	2326	5.6
SRP1848-B09	0.073	97	0.027	74	25152	0.46	25213	0.99	1424	78.0
SRP1848-C05	0.073	92	0.021	62	26836	0.52	15199	0.35	4134	4.8
SRP1848-F02	0.054	92	0.009	54	25714	0.62	14911	0.19	2741	2.6
SRP1848-F08	0.061	94	0.024	77	26483	0.91	21024	1.07	NB	NB
SRP1848-D07	0.075	94	0.032	71	25738	0.77	24272	0.92	NB	NB
SRP1848-F11	0.054	91	0.017	70	26774	0.75	21790	0.47	1762	4.6
SRP1848-F09	0.056	93	0.050	79	23816	0.36	24178	0.75	1671	90.7
SRP1848-D10	0.016	90	0.012	54	26468	0.48	20578	0.52	1859	13.0
SRP1848-G01	0.070	91	0.022	66	27406	0.98	20913	0.56	1993	4.6
SRP1848-B06	0.058	95	0.022	72	25070	0.67	26767	1.21	NB	NB
SRP1848-D03	0.160	98	0.038	76	25977	1.90	14130	0.58	3170	9.5
SRP1848-B07	0.079	96	0.038	73	25612	0.66	25491	1.05	NB	NB
SRP1848-E02	0.046	93	0.025	71	23847	0.53	18717	0.59	1473	21.0
SRP1848-B03	0.050	94	0.028	66	26338	0.82	17228	0.41	2722	6.4

Fab-HC Variant ID	KB, 2° Antibody Cell Killing, Nb-239		Igrov, 2° Antibody Cell Killing, Nb-SC239		CHO-human FolR1		CHO-cyno FolR1		CHO-mouse FolR1	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	Bmax (MFI)	KD (nM)	Bmax (MFI)	KD (nM)	Bmax (MFI)	KD (nM)
SRP1848-E01	0.088	92	0.029	72	26430	1.01	22420	0.96	NB	NB
SRP1848-B05	0.065	94	0.040	72	24536	0.65	21871	0.64	NB	NB
SRP1848-D09	0.042	91	0.023	70	24966	0.46	21306	0.65	NB	NB
SRP1848-F06	0.066	94	0.032	77	25598	0.87	26528	0.86	NB	NB
SRP1848-G10	0.046	97	0.019	79	25269	0.49	14163	0.24	2891	4.3
SRP1848-G04	0.051	92	0.016	75	25156	0.76	12538	0.25	1999	2.5
SRP1848-G06	0.057	96	0.026	81	25838	0.63	12830	0.31	1857	11.1
SRP1848-G07	0.058	94	0.038	78	24939	0.78	13668	0.35	1978	2.9
SRP1848-G09	0.073	97	0.036	83	25066	0.59	17685	0.35	2184	6.4
SRP1848-G11	0.040	97	0.023	84	27191	0.68	11837	0.26	2744	7.6

Table 8. Affinity-matured antibodies from initial leads (Trastuzumab HC framework): Kinetic binding results

Variant ID	Biacore Kinetics		
	ka (1/Ms)	kd (1/s)	KD (M)
SRP1848-A01	8.29E+05	1.55E-03	1.87E-09
SRP1848-A02	5.25E+05	8.82E-03	1.68E-08
SRP1848-A07	1.01E+06	8.66E-04	8.55E-10
SRP1848-C03	1.36E+06	1.52E-03	1.11E-09
SRP1848-F04	8.15E+05	1.08E-03	1.32E-09
SRP1848-B04	7.80E+05	1.17E-03	1.50E-09
SRP1848-B11	1.22E+06	1.86E-03	1.52E-09
SRP1848-F07	1.60E+06	2.49E-03	1.56E-09
SRP1848-E06	9.44E+05	1.54E-03	1.63E-09
SRP1848-A09	7.30E+05	1.33E-03	1.82E-09
SRP1848-E07	1.25E+06	2.40E-03	1.91E-09
SRP1848-G03	9.90E+05	1.97E-03	1.99E-09
SRP1848-A04	1.61E+06	3.26E-03	2.03E-09
SRP1848-H01	6.59E+05	1.39E-03	2.11E-09
SRP1848-B10	6.81E+05	1.48E-03	2.18E-09
SRP1848-C07	8.56E+05	1.89E-03	2.21E-09
SRP1848-F05	6.56E+05	1.57E-03	2.40E-09
SRP1848-D02	8.51E+05	2.05E-03	2.41E-09
SRP1848-A08	4.93E+05	1.19E-03	2.42E-09
SRP1848-E03	6.88E+05	1.83E-03	2.67E-09
SRP1848-A10	1.20E+06	3.30E-03	2.74E-09
SRP1848-F10	8.72E+05	2.47E-03	2.83E-09
SRP1848-D05	6.75E+05	1.98E-03	2.93E-09
SRP1848-C01	7.30E+05	2.23E-03	3.05E-09

Variant ID	Biacore Kinetics		
	ka (1/Ms)	kd (1/s)	K_D (M)
SRP1848-F01	1.14E+06	3.62E-03	3.18E-09
SRP1848-D04	4.97E+05	1.73E-03	3.48E-09
SRP1848-E05	7.16E+05	2.51E-03	3.51E-09
SRP1848-A06	1.37E+06	4.83E-03	3.51E-09
SRP1848-B01	1.13E+06	4.16E-03	3.67E-09
SRP1848-C04	1.29E+06	4.99E-03	3.86E-09
SRP1848-C10	8.99E+05	3.63E-03	4.03E-09
SRP1848-B09	1.55E+06	6.61E-03	4.26E-09
SRP1848-C05	1.06E+06	4.54E-03	4.29E-09
SRP1848-F02	1.42E+06	6.37E-03	4.49E-09
SRP1848-F08	5.94E+05	2.72E-03	4.58E-09
SRP1848-D07	1.09E+06	5.11E-03	4.70E-09
SRP1848-F11	8.28E+05	3.90E-03	4.71E-09
SRP1848-F09	1.40E+06	6.79E-03	4.85E-09
SRP1848-D10	1.13E+06	5.58E-03	4.95E-09
SRP1848-G01	4.44E+05	2.26E-03	5.09E-09
SRP1848-B06	6.20E+05	3.17E-03	5.10E-09
SRP1848-D03	1.03E+06	5.35E-03	5.19E-09
SRP1848-B07	7.06E+05	3.78E-03	5.35E-09
SRP1848-E02	1.14E+06	7.07E-03	6.21E-09
SRP1848-B03	1.13E+06	8.59E-03	7.63E-09
SRP1848-E01	6.64E+05	5.22E-03	7.87E-09
SRP1848-B05	9.76E+05	8.85E-03	9.07E-09
SRP1848-D09	1.07E+06	1.08E-02	1.01E-08
SRP1848-F06	4.56E+05	7.75E-03	1.70E-08
SRP1848-G10	7.58E+05	3.45E-03	4.55E-09

Variant ID	Biacore Kinetics		
	ka (1/Ms)	kd (1/s)	K_D (M)
SRP1848-G04	5.91E+05	3.79E-03	6.40E-09
SRP1848-G06	5.69E+05	3.81E-03	6.70E-09
SRP1848-G07	6.05E+05	4.51E-03	7.45E-09
SRP1848-G09	8.56E+05	6.46E-03	7.56E-09
SRP1848-G11	6.96E+05	6.37E-03	9.14E-09

Table 9. Results obtained with humanized 6D1 (2060) antibodies.

	Biacore kinetics			Igrov 2° AntibodyCell Killing, Nb-SC239	
SRP	**ka (1/Ms)**	**kd (1/s)**	**K_D (M)**	**EC50 (nM)**	**span (%)**
SRP2060-E10	5.82E+05	1.20E-03	2.06E-09	0.061	68
SRP2060-E05	5.41E+05	1.58E-03	2.92E-09	0.22	71
SRP2060-B01	5.61E+05	1.47E-03	2.62E-09	0.045	76
SRP2060-A06	5.47E+05	7.29E-03	1.33E-08	0.013	66

EXAMPLE 8

FOLR1 EXPRESSION IN OVARIAN, ENDOMETRIAL CANCER, NSCLC, AND TNBC CELL LINES

[00563] High levels of FolRα have been found in ovarian and endometrial cancers, triple-negative breast cancer (TNBC) and non-small cell lung carcinoma (NSCLC). Based on reported FolRα mRNA expression levels, a panel of ovarian cancer, endometrial cancer, TNBC, and NSCLC cell lines were selected for *in vitro* testing of candidate ADC molecules. To measure the number of FolRα receptors expressed on the cell surface, Alexa647-conjugated antibody 1848-H01 (Y180/F404) was used in a FACS cell binding assay and FolRα copy number was determined based on Antibody Binding Capacity (ABC) of the conjugated antibody measured by quantitation beads (Simply Cellular anti-human IgG beads from Bangs Laboratories). As shown in Table 10, FolRα copy number on ovarian cancer, endometrial cancer, TNBC, and NSCLC cells ranged from 35,000 to 4,000,000 receptors per cell.

Table 10. FolRα copy numbers in various cell lines

Disease Indication	Cell Line	FolRα Copy # per Cell
Ovarian Cancer	OVKATE	3,590,356
	Igrov1	1,375,828
	OVMANA	1,224,753
	OVSAHO	842,703
	OVISE	678,472
	CAOV3	336,900
	OVCAR3	196,426
	OV90	97,717
Endometrial Cancer	MFE-280	434,941
	HEC-1-A	220,690
	EFE-184	128,166
	HEC-1-B	176,400
	Ishikawa	194,128
	SNG-M	61,961
	NUGC-4	35,395
Lung Cancer	H2342	1,419,355
	H1651	918,800
	H2110	347,447
	H441	251,390
	H226	85,164
	H2405	68,182
	H358	40,058
	H2052	37,677

Disease Indication	Cell Line	FolRα Copy # per Cell
	A549	35,078
	H1770	33,781
TNBC	HCC1143	255,813
	HEC-251	113,270
	HCC1599	65,624
	MDA-MB-468	61,588
	MDA-MB-231	50,005
	HCC38	40,712
	HCC1187	34,936
	HCC1937	23,097

EXAMPLE 9

FOLR1 EXPRESSION IN OVARIAN AND ENDOMETRIAL CANCER, TNBC AND NSCLC TISSUES

[00564] To determine prevalence of FolRα expression in patient samples representative of ovarian and endometrial cancers, TNBC and NSCLC, immunohistochemistry (IHC) staining was performed on commercially available tissue microarrays (TMAs) containing patient samples for the four indications. The TMAs (Biomax; Biomax; Cat# BC11115b, EMC1021, BR1001, and BC041115c) were stained for FolRα expression using a commercial FolRα IHC assay kit (Biocare; **Cat. #IPI4006K G10) with the manufacturer's recommended protocol. Slides were imaged and** stained tumor cores were scored for staining. Positive staining for FolRα was observed in ~80% of ovarian cancer, ~60% of endometrial cancer samples, ~ 30% of TNBC samples and ~ 50% of NSCLC samples; suggesting that these may be suitable indications for a FolRα-targeting therapeutic agent. The relative levels of expression of FolRα in ovarian and endometrial samples is summarized in Table 11.

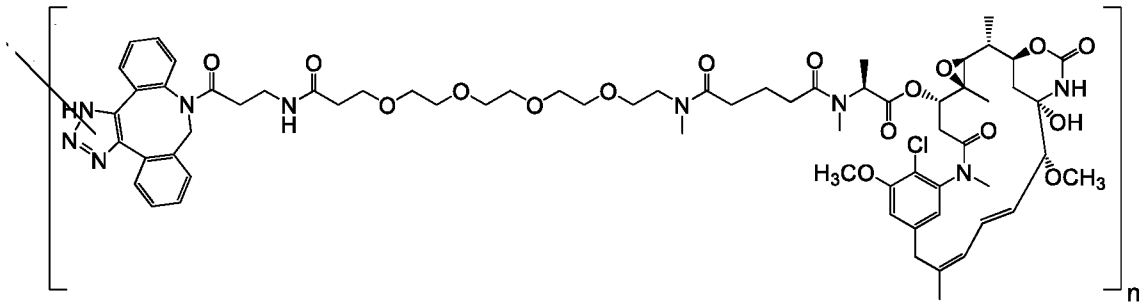
Table 11. Summary of FolRα Expression Levels in Ovarian and Endometrial Cancer Samples

Indication/Cat# of Slide (Biomax US)	Total # of disease cores	Staining intensity			
		0	1+	2+	3+
Ovarian Cancer tissue microarray with adjacent normal tissue, 100 cases/ 100 cores (BC11115b)	97	21	19	29	25
Endometrial Cancer tissue microarray, 102 cases/102 cores (EMC1021)	90	26	37	15	12

EXAMPLE 10

ANTIBODY-DRUG CONJUGATE (ADC) EVALUATION

[00565] Based on biacore affinity to the **extracellular domain of FOLR1 (or "FolRα-ECD"), nine** antibodies were selected for scale-up with para-azidomethyl-L-phenylalanine (pAMF) incorporated at the HC F404 site. The nine antibodies selected for testing were: 1848-A01, 1848-H01, 1848-A08, 1848-B04, 1848-D02, 1848-A07, 1848-B10, 1848-G10, and 1848-G04). Antibody 1848-D02 did not express well and was consequently not used for further investigation. The remaining eight antibodies were conjugated to a non-cleavable maytansine to form antibody-drug conjugates (ADCs) having the structure of **Conjugate M**, below:



Conjugate M

The candidate antibody-**drug conjugates were tested in cell killing on FolRα**-expressing cells, including KB, Igrov1, HeLa and JEG3. Table 12 provides a summary of the *in vitro* cytotoxic activity of the candidate conjugates on KB and JEG3 cells.

Table 12. *In vitro* cytotoxic activity of anti-FOLR1 antibody-drug conjugates

Antibody	[IgG], μg/mL	Killing in KB cells		Killing in JEG3 cells	
		EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
1848-A01	1100	0.16	96	83*	94*
1848-A07	1021	0.17	96	80*	96*
1848-A08	1358	0.17	95	74*	95*
1848-B04	1257	0.3	98	113*	85*
1848-B10	802	0.23	98	43*	82*
1848-D02	1208	Not tested	Not tested	Not tested	Not tested
1848-G04	1415	0.31	94	38*	93*
1848-G10	1746	0.26	92	NC	NC
1848-H01	1723	0.24	93	NC	NC

*Estimated

NC = Not calculable

There was no significant difference between ADCs in cell killing activity on KB and JEG3 cells, Table 12). Accordingly, four leads (1848-A01, 1848-A07, 1848-B04, 1848-G10) conjugated to the non-cleavable maytansine to form structures of **Conjugate M** at a drug-antibody ratio of two (DAR2) were selected for an *in vivo* study based on Biacore affinity (Table 8) and maximizing sequence diversity. Additionally, there was weak cytotoxic activity on JEG3 cells with the ADCs having DAR2.

[00566] To study the effect of drug-antibody ratio (DAR) on the cell killing activity of anti-FolRα leads, the ADCs having DAR2 were also combined with a secondary DAR2 anti-human IgG nanobody conjugated to non-cleavable maytansine to approximate a DAR 4 ADC in cell killing assays. In this assay, 1848-B10 ADC showed the best cell killing activity when combined with the secondary nanobody on JEG3 and Igrov1 cells (data not shown). Based on this result, 1848-B10 ADC at DAR2 was added to an *in vivo* study to evaluate ADC candidates in addition to the other four leads (1848-A01, 1848-A07, 1848-B04, 1848-G10). Results from the *in vivo* efficacy study testing revealed that only 1848-B10 ADC at DAR2 showed weak tumor inhibition in the KB model (data not shown).

[00567] As a result, 1848-B10 was selected as one of the lead antibodies for further ADC studies.

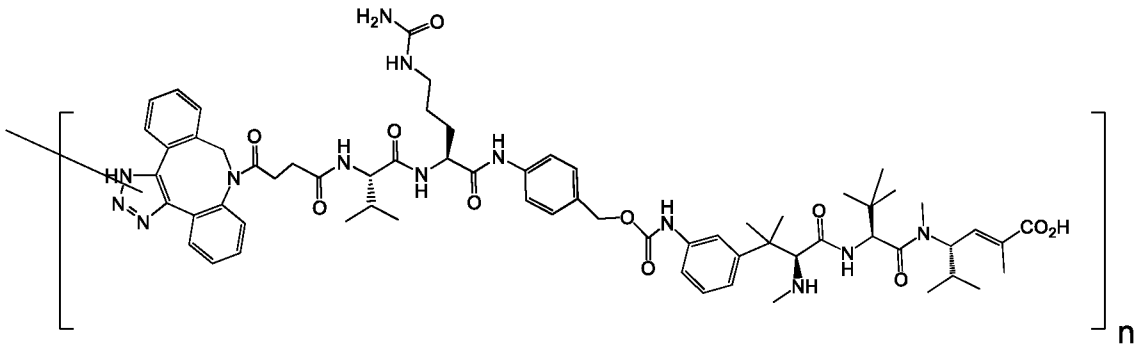
EXAMPLE 11

EFFICACY SCREENING OF TOP FOLR1 ANTIBODY LEADS

[00568] The ADCs containing **Conjugate M** at DAR2 had potent *in vitro* activity against KB cells, which express high levels of FolRα. However, the ADCs had poor *in vitro* activity in JEG3 and Igrov1 cells, which express more moderate levels of FolRα, and low *in vivo* activity in the KB model. The pattern and levels of FolRα expression in JEG3 and Igrov1 cells is more representative of expression in patient tumors, while evaluation of ADC leads in KB cells do not appear to differentiate between the properties of the different leads. FolRα undergoes rapid internalization and recycling without reaching the lysosome; therefore, in order to improve activity of an ADC that targets FolRα, it would be useful to have a linker that can release the drug in the endosomal compartment. Additionally, FolRα expression in primary ovarian tumors and Igrov1 xenografts is heterogenous (Ab *et al.* 2015. *Molecular Cancer Therapeutics* 14(7):1605-1613) suggesting that an ADC with bystander activity could potentially have higher activity in these tumors. To tailor the design of a FolRα-targeting ADC to the biology of the target as well as the expression level and pattern in ovarian cancer, several changes were implemented in the screening strategy. The KB

model was used for primary screening, and the activity of leads was tested on Igrov1 cells *in vitro* and *in vivo* to evaluate and compare the different leads.

[00569] Initial screening of FolRα ADC variants was conducted in KB tumors which express high levels of FolRα. This study sought to evaluate the anti-tumor effects of four different anti-FolRα antibodies conjugated to the same linker-warhead (**Conjugate P**, below) and conjugation sites (Y180/F404). KB cervical carcinoma cells were implanted subcutaneously into athymic nude mice and treated with a single dose of 2.5 mg/kg FolRα ADC variants listed in Table 13. ADC variants were administered when tumors reached ~150 mm^3.



Conjugate P

Table 13. Tested ADC variants

ADC Molecule	Antibody	Conjugation Sites	Conjugate Form	DAR
Vehicle (PBS Only)	--	--	--	NA
1	1848-B10	HC-Y180, F404	P	3.8
2	1848-A07	HC-Y180, F404	P	3.9
3	1848-B04	HC-Y180, F404	P	3.8
4	1848-H01	HC-Y180, F404	P	3.8

[00570] No toxicity was observed with any test article as evidenced by the absence of any significant weight loss, defined as >20% decrease in animal weight (FIG. 6). FIGs. 7 (A, B) illustrate the effects of treatment on KB tumor growth and tumor size on day 25 when the vehicle control group reached the study endpoint (>1000 mm^3). Results show that ADC Molecule 1 (1848-B10 FolRα antibody, Y180/F404, **Conjugate P**) and ADC Molecule 4 (1848-H01 FolRα antibody, Y180/F404, **Conjugate P**) significantly inhibited KB tumor growth compared to control, while the other two ADC variants did not exhibit any anti-tumor activity. By the end of the study on day 31, there was no significant difference between ADC Molecules 1 and 4 (FIG. 8). Therefore,

ADCs containing 1848-B10 and 1848-H01 anti-FolRα antibodies were investigated for further characterization and testing.

EXAMPLE 12

DRUG-ANTIBODY RATIO FOR ANTIBODY-DRUG CONJUGATES

[00571] ADCs with increasing DAR (2-6) and with a cleavable linker were evaluated to determine whether varying these features would improve activity of the molecule. To increase the *in vivo* potency of the FolRα targeting ADCs, 1848-B10 antibodies was expressed with 2, 4, or 6 para-azidomethyl-phenylalanine (pAMF) residues incorporated on each antibody and conjugated to non-cleavable maytansine (**Conjugate M**, Example 10) and cleavable hemiasterlin (**Conjugate P**, Example 11) to generate ADCs with DAR = 2, 4 or 6.

[00572] *In vitro* cell killing on FolRα-positive cells (KB, Igrov1, and JEG3) showed that antibody 1848-B10 conjugates of **Conjugate P** were more potent than 1848-B10 conjugates of **Conjugate M** on Igrov1 cells, which have moderate levels of FolRα expression (Table 14). Additionally, increasing the DAR to 4 resulted in ADCs with greatly improved potency compared to the DAR2 versions, while DAR6 ADCs further improved the cell killing activity only marginally over DAR4. Based on these data, the cleavable hemiasterlin conjugates (**Conjugate P**) at DAR4 was determined to be the optimal conjugate format for the FolRα ADC.

Table 14. Comparison of *in vitro* cytotoxic activity of 1848-B10 ADCs

ADC Molecule	Sites of Conjugation	Linker drug / Conjugate	Expected DAR	Measured DAR	KB (+++)		Igrov (++)		A549 (-)	
					EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
5	F404	Conjugate M	2	1.94	0.74	87	NC	NC	NK	NK
6	K42/F404	Conjugate M	4	3.86	0.42	94	NC	NC	NK	NK
7	K42/Y180/F404	Conjugate M	6	5.54	0.2	96	NC	NC	NK	NK
8	F404	Conjugate P	2	1.89	0.81	80	0.55	56	NK	NK
9	K42/F404	Conjugate P	4	3.69	0.35	94	0.17	62	NK	NK
10	K42/Y180/F404	Conjugate P	6	5.28	0.21	97	0.14	69	NK	NK

NC = Not calculable
NK = No killing

EXAMPLE 13

STUDY TO COMPARE ACTIVITY OF DIFFERENT SITE PAIRS IN ADCS

[00573] This study sought to compare the anti-tumor effects of three different conjugation site pairs (Y180/F404, Y180/K42, and F404/K42) using the same FolRα antibody (1848-B10) and linker-warhead (**Conjugate P**). The *in vitro* cell killing activity of the three ADCs were very similar on KB and Igrov1 cells (Table 15).

Table 15. *In vitro* cell killing activity of tested ADCs (**Conjugate P**)

ADC Molecule	Antibody	Conjugation Sites	Conjugate Form	DAR	EC50 on KB cells (nM)	EC50 on Igrov1 cells (nM)
11	α-GFP	HC-Y180, F404	P	3.58	NK	NK
12	1848-B10	HC-Y180, LC-K42	P	3.93	0.21	0.085
1	1848-B10	HC-Y180, F404	P	3.82	0.21	0.083
9	1848-B10	HC-F404, LC-K42	P	3.90	0.19	0.061

[00574] For *in vivo* efficacy testing, KB cervical carcinoma cells were implanted subcutaneously into athymic nude mice and treated with a single dose of 2.5 mg/kg FolRα ADC variants listed in Table 15. ADCs were administered when tumors reached ~150 mm^3. No toxicity was observed with any test article as evidenced by the absence of any significant weight loss, defined as >20% decrease in animal weight (FIG. 9). FIGs. 10 (A, B) illustrate the effect of treatment on KB tumor growth and tumor size on day 21 when the vehicle treated tumors reached the study endpoint (>1000 mm^3), after which the study was terminated. Results show that all three FolRα ADC variants (ADC Molecules 1, 12, and 9) with different conjugation sites initially induced tumor regression and significantly delayed tumor growth compared to the vehicle control, while control anti-GFP ADC (ADC Molecule 11) behaved similarly to vehicle (FIGs. 10A and 10B). By the end of study on day 36, ADC Molecule 12 exhibited the best duration of response with most tumors in this group remaining growth inhibited, while tumor re-growth was observed for ADC Molecules 1 and 9 (FIG. 10A). Statistical analysis showed that ADC Molecule 12 was significantly more efficacious compared to ADC Molecule 9 (p=0.0297) and ADC Molecule 1 (p=0.0470) (FIG. 11). In conclusion, the Y180/K42 conjugation site resulted in the best potency and duration of response in KB tumors.

EXAMPLE 14

STUDY FOR SELECTION OF LEAD ANTI-FOLR1 ANTIBODIES FOR ADC DESIGN

[00575] To assess potential lead antibodies for anti- FolRα ADCs, a selection of FolRα top leads that were conjugated to form **Conjugate P** with DAR4 were screened *in vitro*. *In vitro* cell

killing activity for the top antibody leads are very similar on KB and Igrov1 cells and, the result is summarized in Table 16.

Table 16. *In vitro* cell killing activity of lead ADCs (**Conjugate P**)

ADC Molecule	Sample	Sites of Conjugation	DAR	KB		Igrov1		A549	
				EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
1	1848-B10	Y180/F404	3.82	0.21	98	0.083	76	NK	NK
2	1848-A07	Y180/F404	3.76	0.18	97	0.084	61	NK	NK
3	1848-B04	Y180/F404	3.84	0.16	97	0.081	68	NK	NK
4	1848-H01	Y180/F404	3.84	0.12	96	0.028	76	NK	NK

[00576] The same **Conjugate P** with DAR4 for the four top lead antibodies were screened in an *in vivo* efficacy study in the KB model (FIGs. 7, 8). Based on results in these studies, 1848-B10 and 1848-H01 were picked as the top antibody leads for further characterization. The sequences of 1848-B10 and 1848-H01, as well as the corresponding CDRs, is shown in Table 32. Additional properties for the top antibody leads are summarized in Table 17.

Table 17. Properties of lead antibodies

Property	**1848-B10, Y180/F404**	**1848-H01, Y180/F404**
K_D (Biacore)	1.4 nM	1 nM
K_D (FACS cell binding, (CHO-h-FOLRα)	4.5 nM	3.7 nM
Cross-reactivity, Cyno (CHO-c-FOLRα)	3.3 nM	3.8 nM
Thermostability (DSC)	66.6°C, 85.9°C	66.8°C, 83.4°C
Mouse PK (ADC, no DAR analysis)*	11.2 days; 6.94 mL/kg/day	14.3 days; 5.46 mL/kg/day
Cyno PK (naked antibody)*	comparable to most antibodies; $T_{1/2}$ = 13.6 days	comparable to most antibodies; $T_{1/2}$ = 8.5 days
ADA in Cyno (naked antibody)*	Very low ADA response	ADA response observed in all animals, affected $T_{1/2}$, no AUC post second dose in one animal
ADC cell killing (Igrov1), Conjugate = P	0.26 nM, 63% span	EC50 < 0.09 nM, span > 70% (averaged across 10 independent experiments)
ADC cell killing (OVCAR3), Conjugate = P		EC50 = 0.03 nM, Span = 71%
Efficacy of multiple leads in KB model and clinical Igrov1 model	Weak tumor inhibition compared to vehicle group (E4)	Significant tumor inhibition compared to vehicle group (E4)

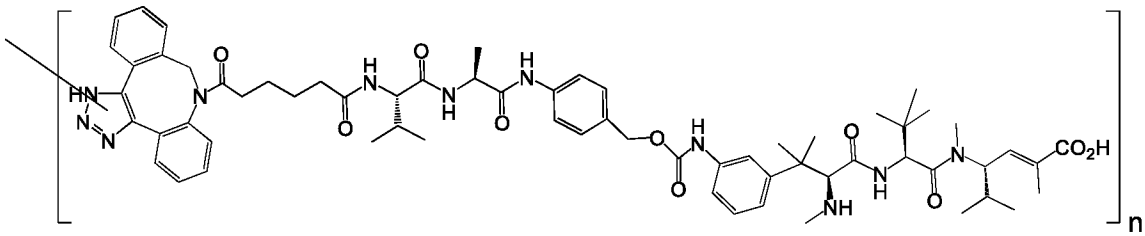
DSC: Differential Scanning Calorimetry

* Surrogate ADCs: (1) 1848-B10, Y180/K42, (2) 1848-H01, Y180/K42

EXAMPLE 15

SELECTION OF OPTIMAL LINKER FOR CLEAVABLE HEMIASTERLIN

[00577] Hemiasterlin with multiple linkers having different cleavage properties were generated. The antibody 1848-B10 was conjugated to several of the candidate linker variants, and the resulting ADCs were tested in *in vitro* cytotoxicity assays (Table 18). Among the candidate linker variants, an alternative of Conjugate P, having a proteolytic sequence of ValAla in place of ValCit, showed good cell killing activity (**Conjugate Q**, below) and afforded a potential advantage in scalability and synthetic efficiency.



Conjugate Q

Table 18. Comparison of *in vitro* cytotoxity of cleavable hemiasterlin linker-drug variants

ADC Molecule	Antibody	Conjugate Form	DAR	Igrov1		A549	
				EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
12	1848-B10, Y180/K42	P	3.74	0.12	78	NK	NK
16	1848-B10, Y180/K42	Q	3.6	0.32	66	NK	NK

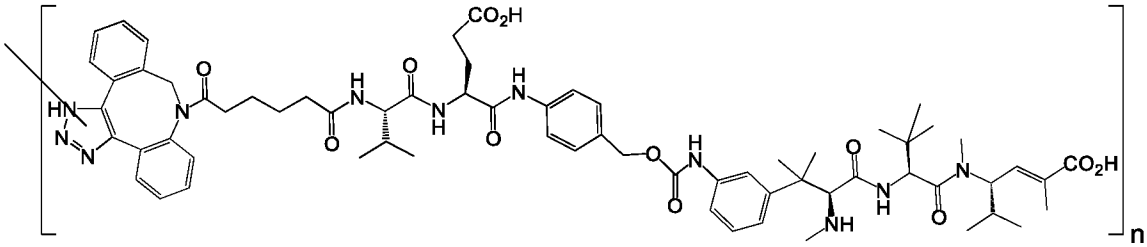
The *in vitro* cytotoxic activity of multiple lots of candidate ADC variants are summarized in Table 19. ADC Molecule 4 showed consistent cell killing with an EC50 ranging from 0.03-0.66 nM and a span ranging from 69-96% across different experiments.

Table 19. Summary of *in vitro* cytotoxicity studies in KB and Igrov1 cell lines

Cell Line	1848-H01, Y180/F404, Conjugate P (ADC Molecule 4)		1848-H01, Y180/F404, Conjugate Q (ADC Molecule 20)		1848-B10, Y180/F404, Conjugate P (ADC Molecule 1)		1848-B10, Y180/F404, Conjugate Q (ADC Molecule 17)	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)

KB	0.66	78			0.45	83		
	0.12	96			0.21	98		
Igrov1	0.11	69			0.26	63		
	0.03	76			0.08	76		
			0.31	72			0.21	71
	0.16	62	0.24	42				
	0.12	68	0.21	53				
	0.06	70	0.13	54				
	0.08	80	0.13	70				
					0.09	81	0.23	70
					0.02	74	0.11	73

[00578] In addition, in a separate study, an antibody-drug conjugate was synthesized using antibody 1848-B10 Y180/F404 and **Conjugate R** (below):



Conjugate R

Conjugate R includes DBCO adipoyl ValGlu linked to a hemiasterlin warhead. The ADC comprising 1848-H01 HC-Y180/F404 with **Conjugate R** (ADC Molecule 22) showed comparable *in vitro* cell-killing activity to ADC Molecule 4 (data not shown) as well as comparable *in vivo* activity in the Igrov1 model (FIGs. 14A, 14B). Accordingly, these results indicate that antibody 1848-B10 HC-Y180/F404 can be used with alternative linker warheads in ADCs targeting FolRα.

EXAMPLE 16

SELECTION OF AN OPTIMAL ADC LEAD

[00579] This study sought to assess different aspects of the FolRα ADC molecule including the antibody, conjugation sites, and linker warhead. Igrov1 ovarian cancer cells were implanted subcutaneously into SCID Beige mice and treated with a single dose of 2.5 mg/kg FolRα ADC variants listed in Table 20. ADCs were administered when tumors reached ~150 mm^3.

Table 20. ADCs tested in the efficacy screening

ADC Molecule	Antibody	Conjugation Sites	Conjugate Form	DAR	Igrov1 Cell Killing	
					EC50 (nM)	Span (%)
1	1848-B10	Y180/F404	P	3.82	0.09	76
17	1848-B10	Y180/F404	Q	3.74	0.21	71

12	1848-B10	K42/Y180	P	3.72	0.12	78
16	1848-B10	K42/Y180	Q	3.6	0.32	66
4	1848-H01	Y180/F404	P	3.84	0.03	76
18	1848-H01	K42/Y180	P	3.87	0.14	70
19	1848-H01	K42/Y180	Q	3.61	0.23	52

[00580] No toxicity was observed with any test article as evidenced by the absence of any significant weight loss, defined as >20% decrease in animal weight (FIG. 12). FIG. 13 (A, B) illustrates the effects of treatment on Igrov1 tumor growth and final tumor size on post treatment day 24 when the vehicle control treated tumors reached the study endpoint (~1000 mm^3). Out of seven FolRα ADC variants tested, ADC Molecule 4 significantly inhibited tumor growth compared to the vehicle control (FIGs. 13A,13B). This result identifies 1848-H01, Y180/F404, and SC239 as an optimal combination of anti-FolRα antibody, conjugation sites, and linker warhead, respectively, in Igrov-1 tumors.

EXAMPLE 17

CROSS-REACTIVITY OF ADC VARIANTS TO FOLR ISOFORMS

[00581] The folate receptor has three isoforms in humans, termed hFolRα, hFolRβ, and hFolRγ (also FOLR1, FOLR2, and FOLR3, respectively). hFolRα and hFolRβ are expressed at the plasma membrane via a GPI anchor, whereas FolRγ is secreted. In normal tissues FolRα is generally expressed on the apical surface of polarized epithelial cells, while hFolRβ is expressed in latter stages of normal myelopoiesis and in the placenta, spleen, and thymus. In the normal development of the myelomonocytic lineage, hFolRβ is seen as a differentiation marker coexpressed with CD14 at relatively low levels in monocytes but not in CD34+ normal hematopoietic progenitors. hFolRγ is secreted at low levels from lymphoid cells in the spleen, thymus, and bone marrow. The three FR isoforms have a high degree of homology with FolRα, sharing 72% and 71% sequence identity with FolRβ and FolRγ respectively. Therefore, it is useful to determine the specificity of lead antibodies 1848-B10 and 1848-H01 for FolRα, and the extent of cross reactivity with cells expressing FolRβ and FolRγ.

[00582] 293T cells stably expressing the three folate receptor isoforms (hFolRα, hFolRβ, and hFolRγ) were generated and tested for binding to antibodies 1848-B10 and 1848-H01 in a FACS assay. In this assay, 1848-H01 but not 1848-B10 showed very weak binding to FolRβ (FIG. 15) but not to FolRγ (not shown). The 1848-H01 binding to FolRβ expressing cells had an affinity of 156 nM, with a B_{max} that was only 20% compared to the B_{max} for FolRα. Assessment of the cytotoxic activity of the corresponding ADC Molecules 4 and 1 on 293T cells expressing the FolRα and FolRβ isoforms showed that ADC Molecule 4 had a weak but specific cytotoxic effect

on cells expressing FolRβ at concentrations above 10 nM, with an EC50 of ~100 nM compared to an EC50 of < 10 nM for FolRα expressing cells (FIG. 16).

EXAMPLE 18

BINDING AND CYTOTOXIC ACTIVITY OF ADC VARIANTS IN HEMATOPOIETIC CELLS

[00583] To determine if ADC Molecules 1 and 4 have an effect on viability of hematopoietic cells, FolR expression was determined in T cells, B cells and monocytes in isolated PBMCs (n=4 donors) and the extent of antibody 1848-H01 and 1848-B10 binding to FolRβ on immune cells was assessed. Heterogenous (donor variable) FolRα expression was detected in monocytes but this expression was transient and disappeared after 1 day in culture (data not shown), whereas FolRβ was consistently expressed in a subpopulation of monocytes (data not shown). Neither antibody 1848-B10 nor 1848-H01 was observed to bind monocytes, although FolRβ expression was detectable on these cells (data not shown). Further, CD14 monocytes were assayed for viability following treatment with ADC Molecules 1 and 4 and to address potential cytotoxicity. In correlation with negative cell binding, the ADC variants did not affect viability of monocytes/macrophages, suggesting no clinical impact on PB monocytes in humans (data not shown).

[00584] FolRβ is weakly expressed on M1 macrophages, and highly expressed on M2 macrophages and their subsets. Antibodies 1848-H01 and 1848-B10 were therefore assessed for their ability to bind isolated macrophages. However, neither antibody showed any binding to M1 or M2 macrophages although FolRβ expression was confirmed in these cells. To confirm this lack of interaction, the corresponding ADC molecules (1, 4) were assessed for cell killing activities on polarized macrophages (10,000 cells, incubation period = 3 days). Consistent with the lack of binding, neither ADC variant showed any cytotoxic activity on the macrophages from multiple donors (data not shown).

[00585] Accordingly, the ADC variants were demonstrated to have minimum binding and cytotoxic impact on monocytes and macrophages isolated from human donors.

EXAMPLE 19

ADDITIONAL CHARACTERIZATION OF ANTIBODY-DRUG CONJUGATES

[00586] Lead antibody-drug conjugates (ADCs) comprising an anti-FolRα antibody were evaluated and characterized. The characteristics that were measured and analyzed included expression and purification profiles of the lead ADCs, conjugation efficiency and *in vitro* and *in*

vivo activity of the ADC in clinically relevant models. The properties of the anti-FolRα lead ADCs are summarized in Table 21.

Table 21. Properties of lead anti-FolRα ADCs

Property	ADC Molecule 4	ADC Molecule 20
DAR by MALDI	3.73	3.81
Conjugation Efficiency	93%	95%
ADC cell killing	Igrov1: EC50 = 0.08nM. span = 80%	Igrov1: EC50 = 0.13 nM, span = 70%
Preclinical *in vivo* efficacy	Complete tumor growh inhibition at 5 mpk dose and above	Complete tumor inhibition at 10 and 15 mpk, weak tumor inhibition at 5 mpk

EXAMPLE 20

DOSE RANGING EFFICACY STUDY

[00587] The dose-response relationship of ADC Molecules 4 and 20 was evaluated in Igrov-1 tumors. This study sought to: (1) assess which FolRα ADC variant conjugated to hemiasterlin-based linker warheads (Conjugates P, Q) was superior; (2) compare the anti-tumor activity of these FolRα ADC variants to a comparator molecule (ADC Molecule 21), and (3) determine the minimum and maximum efficacious dose of the more efficacious FolRα ADC variant identified. All test articles are decribed in Table 22.

Table 22. ADCs tested in the dose range study

ADC Molecule	Antibody	Conjugation Sites	Conjugate Form	DAR
Vehicle (PBS Only)	NA	NA	NA	NA
4	1848-H01	Y180/F404	P	3.73
20	1848-H01	Y180/F404	Q	3.76
21	Mov19-sulfo-SPDB-DM4			3.3

[00588] SCID Beige mice with established Igrov1 ovarian tumors were treated once with 4 doses of ADC Molecule 4 or 20, with the dosage ranging from 2.5 mg/kg to 15 mg/kg. For comparison, the benchmark group was treated once with 5 mg/kg of a comparator ADC molecule (ADC Molecule 21). No toxicity was observed with any test article as evidenced by the absence of significant weight loss (defined as >20% decrease in animal weight) (FIG. 17). FIG. 18 (A, B, C) illustrates the effects of treatment on Igrov1 tumor growth and the individual tumor sizes until post treatment day 21 when the vehicle control treated tumors reached the study endpoint (> 1000 mm^3). Comparison of tumor size on day 21 (versus vehicle control) indicates that 5

mg/kg and 10 mg/kg doses of ADC Molecule 4 are more efficacious than equivalent doses of ADC Molecule 20 or comparator ADC Molecule 21 based on lower p values (FIG. 18C). At the highest dose (15 mg/kg), both ADC Molecules 4 and 20 demonstrated potent anti-tumor activity with similar p values compared to vehicle control (FIG. 18C). Side by side comparison of tumor growth curves sorted by dose revealed that ADC Molecule 4 was more potent than ADC Molecule 20 based on superior activity of ADC Molecule 4 at lower doses (FIG. 19, A-D). Tumor stasis was observed until day 26 post treatment at 5 mg/kg ADC Molecule 4 versus at 10 mg/kg ADC Molecule 20 (FIGs. 19B, 19C). Tumor regression was induced starting at 10 mg/kg ADC Molecule 4 versus 15 mg/kg ADC Molecule 20 (FIGs. 19C, 19D). In addition, ADC Molecule 4 significantly delayed tumor growth to reach 300 mm^3 compared to ADC Molecule 20 at 5, 10 and 15 mg/kg and comparator ADC Molecule 21 at 5 mg/kg (FIG. 20).

[00589] Cumulatively, these results demonstrate that ADC Molecule 4 is significantly more more efficacious than ADC Molecule 20 and ADC Molecule 21 in Igrov1 tumors. The minimum efficacious dose of ADC Molecule 4 was observed at 5 mg/kg, while 15 mg/kg was the maximum efficacious dose with longest duration of response.

EXAMPLE 21
EFFICACY OF ADC VARIANTS IN COMBINATION TREATMENT WITH CARBOPLATIN

[00590] The efficacy of ADC Molecule 4 in combination with a standard chemotherapeutic agent for ovarian cancer, carboplatin, was evaluated in Igrov1 tumors. Animals bearing established Igrov1 tumors (average tumor size 150 mm^3) were treated with a single dose of 2.5 mg/kg ADC Molecule 4 with or without 60 mg/kg carboplatin every 7 days, for two treatments (q7dx2). FIG. 21A illustrates the effects of treatment on Igrov1 tumor growth until post treatment day 29 when the mean of vehicle control treated tumors reached the study endpoint (~1200 mm^3). Analysis of final tumor size and tumor growth inhibition (TGI) on day 29 showed that single agent ADC Molecule 4 and carboplatin exhibited moderate activity compared to vehicle control with TGI ranging from 50% and 70%, respectively (FIGs. 21B and 21C). The combination of ADC Molecule 4 with carboplatin significantly improved efficacy compared to carboplatin alone, but the combination was not significantly different compared to the single agent ADC Molecule 4 (FIG. 21A). The final mean tumor size in combination treated animals was significantly smaller compared to single agent carboplatin treated animals (414 mm^3

vs. 842 mm^3, p = 0.0011) (FIG. 21B). In addition, TGI in combination treated group was higher at 79% vs. 47% for single agent carboplatin group (p = 0.0008) (FIG. 21C).

[00591] In conclusion, significant added benefit was observed when ADC Molecule 4 was combined with carboplatin compared to single agent carboplatin. This observation was consistently reproduced in two additional independent studies using the same model dosed with similar doses of ADC Molecule 4 and carboplatin (data not shown).

EXAMPLE 22

EFFICACY OF ADC VARIANTS IN OVARIAN TUMOR MODELS

[00592] The efficacy of ADC Molecule 4 was evaluated in human ovarian cell line OVCAR3 tumor models. Animals bearing established OVCAR3 tumors ranging from 100 – 200 mm^3 were treated with a single dose of 2.5 or 5 mg/kg of ADC Molecule 4. FIG. 22 (A, B) illustrates the effects of treatment on OVCAR3 tumor growth and final tumor size on post treatment day 31 when the mean of vehicle control treated tumors reached >1500 mm^3. Treatment with 2.5 mg/kg ADC Molecule 4 resulted in tumor stasis until around 12 days post treatment, while 5 mg/kg ADC Molecule 4 induced tumor regression with regrowth observed around day 20 post treatment (FIG. 22A). Analysis of final tumor size on day 31 showed that treatment with 2.5 and 5 mg/kg of ADC Molecule 4 were both significantly efficacious compared to vehicle control exhibiting 60% and 89% tumor growth inhibition (TGI), respectively (FIG. 22B).

EXAMPLE 23

EFFICACY OF ADC VARIANTS IN ENDOMETRIUM PATIENT DERIVED XENOGRAFT MODELS

[00593] Endometrium cancer patient derived xenograft (PDX) models were assessed for FolRα expression levels by immunohistochemistry analysis of xenograft tissue using a biotinylated mouse monoclonal antibody against FolRα. The efficacy of ADC Molecule 4was assessed in a subset of these PDX models and included models with negative, low (+), medium (++) and high (+++) FolRα expression. Animals bearing established (~ 100-200 mm^3) PDX tumors received 10 mg/kg ADC Molecule 4 weekly via intravenous (IV) injection (n=3) or no treatment (control, n=2-3) until the group mean was >1,000 mm^3 or until day 45 post treatment. If the tumors reached 1,000 mm^3 before Day 14 post treatment, the endpoint was extended to 2,000 mm^3.

[00594] Statistically significant efficacy was observed in about 50% of the FolRα positive models tested, with tumor growth inhibition (TGI) ranging from approximately 50% to greater than 100 % (indicating regression below the tumor size at the start of treatment).

Meanwhile, no significant activity was observed in all PDX models with negative FolRα expression. The degree of anti-tumor activity of ADC Molecule 4 appeared to positively correlate with FolRα expression levels (e.g. PDX tumors with higher levels of FolRα exhibited higher TGI in response to treatment with ADC Molecule 4). The data shown (FIG. 23) is a representation of some of the models that exhibited efficacy: (A) PDX model with FolRα–negative expression; (B) PDX model with FolRα+ expression; (C, D) PDX models with FolRα++ expression; (E, F) PDX models with FolRα+++ expression. Percent TGI (determined on the last day of control tumors) and corresponding p values are indicated on the graphs. Statistical analysis of TGI was performed using an unpaired t test. A probability of less than 5% ($p < 0.05$) was considered as significant. All graphs are presented as mean ± SEM.

EXAMPLE 24

EFFICACY OF ADC VARIANTS IN COMBINATION TREATMENT WITH AVELUMAB

[00595] The efficacy of ADC Molecule 4 in combination with PD-L1 inhibitor Avelumab (clinical grade) was evaluated in animals bearing established MC38-FolRα tumors. Results are illustrated in FIGs. 24 and 25. FIGs. 24A and 25A illustrate MC38-hFolRα tumor growth curves in response to indicated doses of ADC Molecule 4, Avelumab, or a combination of both. Growth curves are shown until > 50% of animals in the single agent treatment groups were euthanized due to reaching tumor size limit based on IACUC protocol. FIG. 24B is a scatter plot of individual tumor size on day 12 when mean of control tumors was > 1,200 mm^3. Statistical analysis for comparison to vehicle control was performed using one-way ANOVA with **Dunnett's multiple comparisons test. A probability of less than 5% ($p < 0.05$) was considered as** significant. FIG. 25B is a Kaplan-Meier curve that shows the fraction of animals that survive in response to treatment with indicated doses of ADC Molecule 4, Avelumab or combination of both. All graphs are presented as mean or individual values ± SEM.

[00596] As illustrated in FIG. 24, single agent ADC Molecule 4 at either dose (10 mg/kg or 15 mg/kg once via IV injection) or Avelumab (administered q3dx3 intraperotineally) initially resulted in tumor stasis until approximately day 7, while combination treatment induced tumor regression (FIG. 24A). Analysis of tumor size on day 12 showed significant inhibition of tumor growth in all treatment groups compared to vehicle control (FIG. 24B). Continued monitoring revealed that ADC Molecule 4 in combination with Avelumab markedly enhanced anti-tumor activity compared to either single agent alone as evidenced by complete regression (e.g. no palpable tumors) in 14 out of 15 animals (FIG. 24B).

[00597] As illustrated in FIG. 25, tumor re-growth was observed in one animal in the 10 mg/kg ADC Molecule 4 + Avelumab group and was euthanized on day 59 due to reaching the maximum tumor size (FIG. 25A). Furthermore, combination treatment demonstrated curative effects or complete remission based on significantly prolonged survival of healthy animals with normal body weight gain and no tumor re-growth up to 112 days post-treatment which is 3-4 fold longer median survival than single agents (FIG. 25B).

EXAMPLE 25

PHARMACOKINETIC PROPERTIES OF ADC VARIANTS IN SCID BEIGE MICE

[00598] The non-compartmental pharmacokinetic parameters of candidate FolRα ADC variants that demonstrated good efficacy in the KB and Igrov1 tumor models were evaluated in non-tumor bearing SCID Beige mice. A single 5 mg/kg IV bolus was administered, sampled and pooled from different mice to obtain time-points for pharmacokinetic (PK) parameters. FolRα ADC variants do not bind murine FolRα, therefore, antigen-mediated PK effects are not expected. A list of tested articles and summary of the results are presented in Table 23.

Table 23. Pharmacokinetic parameters of FolRα ADC variants in SCID Beige mice

Parameters	Units	ADC Molecule 4	ADC Molecule 20	ADC Molecule 21
Dose	mg/kg	5	5	5
Study length	Days	21	21	21
$T_{1/2}$	Days	6.36	5.48	7.59
C_0	µg/mL	122	125	115
Cmax ± SE	µg/mL	118 ± 5	123 ± 10	113 ± 4
$AUC_{(0\text{-all})}$ ± SEM	day* µg/mL	476 ± 22	543 ± 22	447 ± 8
$AUC_{(0\text{-}\infty)}$	day* µg/mL	523	580	510
CL	mL/day/kg	9.57	8.63	9.8
V_{SS}	mL/kg	79.7	62.2	95.2

[00599] The elimination half-life ($T_{1/2}$) was determined from a regression analysis of the log-linear plot of the concentration-time curves. Specifically, $T_{1/2}$, CL and Vss of ADC Molecule 4 were 6.36, 9.57 and 79.7, respectively. In addition, the C_{max} for ADC Molecule 4 was determined to be 118 ± 5 µg/mL.

[00600] In general, the pharmacokinetic properties of all FolRα ADC variants tested were comparable and exhibited similar PK profiles values (FIG. 26). In addition, the murine pharmacokinetic profile of all test articles exhibited PK profiles that are similar those of other FDA-approved monoclonal IgG antibodies.

EXAMPLE 26

PHARMACOKINETIC PROPERTIES OF ADC LEAD ANTIBODIES IN CYNOMOLGOUS MONKEYS

[00601] The non-compartmental pharmacokinetic (PK) parameters of antibodies 1848-B10 and 1848-H01 with K42/Y180 conjugation sites was assessed in cynomologous monkeys (n=3 for each antibody dose) in a repeat dose study. Two 10 mg/kg IV doses were administered on day 1 and day 15, and samples analyzed to determine PK parameters and anti-drug antibody (ADA) response. The two antibodies bind cyno FolRα with comparable affinity to the human target, therefore, antigen-mediated PK effects would be expected. As summarized in Table 24, PK profiles for both antibodies are similar.

Table 24. Pharmacokinetic parameters of lead anti-FolRα antibodies in cynomolgous monkeys

Treatment		Terminal $t_{1/2}$	C_0	$AUC_{(0\text{-last})}$	$AUC_{(0\text{-}\infty)}$	Clearance	Vss
		(day)	(μg/mL)	(day* μg/mL)	(day* μg/mL)	(mL/day/kg)	(mL/kg)
1848-B10, K42/Y180 (dose 1)	Mean	9.73	263	1100	1700	6.02	78.8
	SE	1.01	8	95	171	0.59	8.4
1848-B10, K42/Y180 (dose 2)	Mean	13.1	241	1670	2110	4.74	83.2
	SE	2.3	14	120	40	0.09	15.5
1848-H01, K42/Y180 (dose 1)	Mean	6.54	267	1010	1310	7.83	67.3
	SE	0.51	8	60	130	0.83	2.0
1848-H01, K42/Y180 (dose 2)	Mean	8.08	220	1370	1530	6.60	71.3
	SE	2.51	25	30	110	0.47	12.1

[00602] Mean pharmacokinetic parameters for antibody 1848-H01 (K42/Y180) were similar after Doses 1 and 2. Mean plasma clearance after Doses 1 and 2 was 7.83 and 6.60 mL/day/kg, respectively, and distribution volume was 67 and 71 mL/kg, respectively. Mean terminal half-life for Doses 1 and 2 was 6.5 and 8.0 days, respectively. Mean pharmacokinetic parameters for 1848-B10 (K42/Y180) were comparable after Dose 1 and Dose 2. Plasma clearances after Doses 1 and 2 was 6.02 and 4.74 mL/day/kg, respectively. Mean terminal half-life values for Doses 1 and 2 were 9.7 and 13 days, respectively. Distribution volume was approximately 80 mL/kg.

[00603] Serum samples from the treated animals were also analyzed for the development of anti-drug antibodies (data not shown). ADA analysis showed no significant response at day 15 (post 1st dose) for both antibodies, however ADA was detected in several animals at day 28 and 43 (post 2nd dose at day 15).

EXAMPLE 27

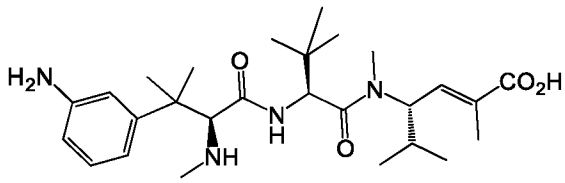
PHARMACOKINETIC EVALUATION OF AN ADC CANDIDATE IN CYNOMOLGOUS MONKEYS

[00604] Female cynomolgus monkeys were administered IV slow bolus doses of vehicle control or ADC Molecule 4 at doses of 1, 3, 10 and 30 mg/kg on Days 1 and 22 (n=3/group) and were observed until day 43. Serum and plasma were collected at several time points from all groups for toxicokinetic profile evaluation (total antibody, ADC, and free drug (I) catabolite). Toxicokinetic analysis confirmed exposures of ADC Molecule 4 at all doses, assessed by evaluating circulating levels of the ADC, total antibody, and free drug (I). The mean C_{max} and AUC values of ADC, total antibody, and free drug (I) increased with increases in dose levels of ADC Molecule 4 in an approximately dose proportional manner and were generally similar on Days 1 and 22. The half-life ($T_{1/2}$) of the ADC ranged from 1.7 to greater than 2 days, and C_{max} ranged from 29-560 μg/mL depending on the dose administered.

EXAMPLE 28

IDENTIFICATION OF CATABOLITES RELEASED FROM ADC CANDIDATES

[00605] The anti-FolRα ADCs described here are predicted to be processed within the endosome or lysosome resulting in release of the metabolite, free drug (I), that may permeate surrounding cells and can cause bystander activity. The free drug released from conjugates P and Q is predicted to be a compound of structure (I), which is illustrated below:



(I)

[00606] The generation of free drug (I) was confirmed in cultured cells (not shown) and Igrov1 tumors treated with ADC Molecule 1 and ADC Molecule 17 with tumors harvested at different timepoints post dosing. Tumors were homogenized and extracted in acetonitrile and the solvent extracted fraction was analyzed by LC/MS. In the animals treated with ADC Molecule 1 and ADC molecule 17, catabolite C1 was found in the tumor samples but not in the plasma of the treated mice (FIG. 27). The LC/MS profile of C1 matched that of the predicted catabolite free drug (I) and the structure was confirmed by mass spectrophotometric analysis (not shown). Free drug (I) was shown to have cytotoxic activity *in vitro,* with an EC_{50} ranging from 0.5-20 nM depending on the cell line tested (Table 26, data for all cell lines not shown).

EXAMPLE 29

IN VIVO STABILITY OF ADC CANDIDATES

[00607] The *in vivo* stability of ADC Molecule 4 was measured in nude strain of mice following a single dose of ADC at 5 mg/kg. Plasma was collected at various time points and analyzed for total IgG by ELISA. The DAR analysis of circulating ADC was measured by affinity capture followed by LC-MS. Data shows that the DAR value does not change over the course of the study FIG. 28. The observed degradation peak also does not change during the run and is present in a similar amount as see in the original stock.

EXAMPLE 30

STABILITY OF ADC CANDIDATES IN PLASMA

[00608] The stability of ADC Molecules 4 and 20 was tested in cynomolgous and human plasma to compare stability of the two linker-drugs, Conjugates P and Q. The ADCs were incubated in duplicate in PBS, human and cynomolgous plasma at 50 μg/mL. The samples were incubated for time points from 60 min, 1 day, 3 days, 7 days, 14 days, and 21 days. Total IgG by ELISA and DAR analysis of the ADC with affinity capture and LC-MS was used to assess the stability of the molecule. Data shown in FIG. 29 shows that the *in vitro* stability of ADC Molecules 4 and 20 was comparable in both human and cyno plasma, with DAR4 being retained until day 21. Both molecules also showed occurrence of clipping, likely in the C-terminal end of the antibody, which was observed starting at day 1 post incubation. This clipping is likely to be cleavage of the two lysine residues at the C-terminal end of the heavy chain and is unlikely to impact stability or activity of the molecule. Similar clipping is commonly observed during CHO production of IgG molecules.

EXAMPLE 31

COMPARISON OF ADC CANDIDATE TO A COMPARATOR ADC

[00609] A comparator of the ADC candidates described herein is IMGN853. IMGN853 (mirvetuximab soravtansine) is an antibody-drug conjugate containing a FolRα-binding antibody linked to the tubulin-disrupting maytansinoid, DM4, via a cleavable (sulfo-SPDB) linker. The design of IMGN853, including selection of its antibody and linker components, was based on optimization of its antitumor activity in preclinical models having levels of FolRα expression representative of those in tumor samples from patients with ovarian and non-small cell lung cancer. Although IMGN853 looks promising in the clinic, based on its chemistry, it may have some potential liabilities that affect the stability, safety and activity of the molecule.

Accordingly, assays to evaluate the properties and pre-clinical effects of an IMGN853 surrogate (ADC Molecule 21) and ADC Molecule 4 are described below.

[00610] To assess the specificity of ADC Molecule 4 compared to that of IMGN853, the cytotoxic activities of ADC Molecule 4 was compared to a closely-approximating surrogate for IMGN853. The surrogate was expressed transiently in CHO cells and conjugated to sulfo-SPDB-DM4 to produce ADC Molecule 21. Cytotoxic activities of the two ADC molecules were compared in the presence of excess un-conjugated "naked" antibody as competitior, on cells that were positive FolRα expression (Igrov1 and OVCAR3) and on cells that were negative for FolRα expression (A549). For ADC Molecule 4, the cell killing activity on Igrov1 cells was reduced by about 800-fold in the presence of un-conjugated antibody (from an EC50 of 0.053 nM to an EC50 greater than 33 nM), indicating that the cell killing activity of ADC Molecule 4 is specific to the presence of FolRα antigen on the cell surface, since the naked antibody competes with the ADC for binding to the FolRα antigen. Cell killing activity of ADC Molecule 21 is not completely dependent on the presence of FolRα antigen on the cell surface since the addition of naked antibody only shifted the EC50 by about 3-fold on Igrov1 cells. Similar results were also observed on OVCAR3 cells (FIGs. 30A, 30B). On FolRα negative A549 cells, potent non-specific cell killing was observed for ADC Molecule 21, but non-specific cell killing was not observed for ADC Molecule 4 (FIG. 30C). Based on these data, it can be concluded that that ADC Molecule 4 shows potent and specific cell killing only on FolRα positive cells, while ADC Molecule 21 shows non-specific cell killing that is not related to ADC binding to FolRα antigen. The results are summarized in Table 25.

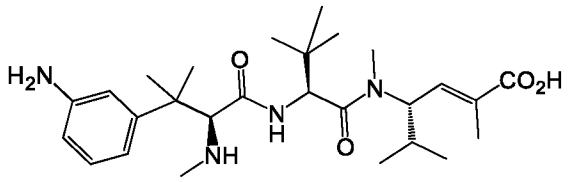
Table 25. Specific cytotoxic activities of ADC Molecule 4 and ADC Molecule 21 in FolRα positive and negative cells

Sample Tested	Igrov1		OVCAR3		A549	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
ADC Molecule 4	0.053	66	0.58	53	NK	NK
ADC Molecule 4 + 0.5uM 1848-H01	>33	NC	>33	NC	NK	NK
ADC Molecule 21	3.9	87	7.9	100	7.4	79
ADC Molecule 21 + 0.5uM Mov19	~ 11	80	10	99	7	81

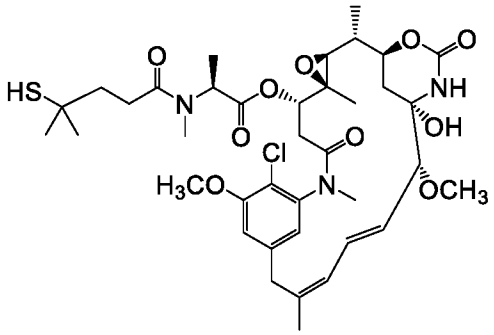
NC = Not Calculable
NK = No Killing

[00611] ADC Molecule 4 had specific cell killing activity on FolRα-positive Igrov1 cells but did not have any activity on A549 FolRα-negative cells. In contrast, ADC Molecule 21 had cytotoxic activity on both negative- and positive-FolRα cell lines, suggesting a lack of specificity

that can be attributed to the potential instability of the sulfo-SPDB linker under reducing conditions in culture and *in vivo* or due to pinocytosis of the ADC into the cells. The free drugs released from ADC Molecule 4 and ADC Molecule 21 are compounds of structure (I) and (II), respectively, which are illustrated below:



(I)



(II)

In *in vitro* cytotoxicity studies, the released free drugs (I) and (II) had comparable cytotoxic activity (Table 26). One key observation in this study is that ADC Molecule 4 is 10-fold more potent than the theoretical four free drug moieties (I) that it is conjugated to, indicating that the conjugate confers higher levels of specific killing to target cells than free drug (I).

Table 26. Cytotoxic activities in Igrov1 and A549 cells

Molecule	Igrov1		A549	
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)
ADC Molecule 4	0.06	70	NK	NK
ADC Molecule 21	4.4*	85	9*	71
Free drug (I)	2.4	92	11	81
Free drug (II)	4.9	90	7.7	81

*estimated EC50 based on incomplete titration
NC = Not Calculable
NK = No Killing

[00612] To assess the pharmacokinetic profile of free drug (I), female Sprague-Dawley rats (mean body weight 250 g) were given either a 0.4 mg/kg or 1 mg/kg dose by IV bolus administration (N= three animal per dose level) via an indwelling jugular vein catheter. Blood samples were collected at 0, 0.83, 1, 2, 8, 24, 32, 48, and 72 hour post-dose. Levels of free drug (I) and (II) were measured by LC-MS/MS and non-compartmental pharmacokinetic analysis was

conducted using Phoenix WinNonlin Version 6.4 (Pharsight Corporation). Table 27 provides the PK data collected from this study. The PK of free drug (I) could not be estimated due to the fact that concentrations of (I) increased from 2 to 8 hours and were undetectable thereafter. The results of this study suggest that clearance of free drug (I) is more rapid than that of free drug (II) since (I) is undetectable at 24 hours post dose (data not shown). In addition, administration of the two doses of free drug (I) did not have an effect on the body weight of the animals, while progressive body weight loss was observed upon treatment with 0.4 mg/kg of free drug (II) (FIG. 31).

Table 27. Pharmacokinetic data for free drugs (I) and (II)

Compound ID	Dose Level	Terminal $t_{1/2}$ (hr)	C_{max} ± SE (ng/mL)	C_0 (ng/mL)	$AUC_{0\text{-last}}$ ± SE (hr*ng/mL)	$AUC_{0\text{-inf}}$ (hr*ng/mL)	Clearance (L/hr/kg)	V_{SS} (L/kg)
Free drug (I)	0.1 mg/kg	Insuff. data	5.82 ± 0.69	6.81	11.0 ± 2.1	Insuff. data	Insuff. data	Insuff. data
Free drug (I)	0.4 mg/kg	Insuff. Data	12.8 ± 2.1	15.5	23.9 ± 2.6	Insuff. data	Insuff. data	Insuff. data
Free drug (II)	0.1 mg/kg	22.4	9.31 ± 2.19	10.7	96.9 ± 11.0	129	0.775	15.8
Free drug (II)	0.4 mg/kg	44.3	53.3 ± 19	62.5	211 ± 70	242	1.66	44.1

[00613] Table 28 is a summary of the properties of ADC Molecule 4 and its metabolite, free drug (I).

Table 28. Properties of ADC Molecule 4 and free drug (I)

Property/Characteristic	Results for ADC Molecule 4
PK of ADC Molecule 4 in mouse with DAR analysis	$T_{1/2}$: 6.38 days; Clearance rate ~ 9.5 mL/kg/day; DAR4 retained to day 21
Stability of ADC Molecule 4 in human and cyno plasma with DAR analysis	DAR4 retained to day 21
PK of free drug (I) in rat vs. free drug (II)	Free drug (I) has faster clearance than (II)
Specificity of activity	ADC Molecule 4 is not active in cell lines that do not express FolRα

EXAMPLE 32

COMPARISON OF STABILITY OF DRUG LINKAGE IN ADC CANDIDATE VS. A COMPARATOR ADC

[00614] The stability of ADC Molecule 4 and comparator ADC Molecule 21 was assessed in cynomolgous monkey and human plasma and PBS, followed by quantitation of the released catabolites, free drugs (I) and (II), respectively (*see* Example 29). Based on the data summarized in FIG. 32, the drug linkage for ADC Molecule 4 appears to be more stable than that for ADC Molecule 21. ADC Molecule 21 appears to be rapidly cleaved in human and cyno plasma, such that the free drug (II) is detected within 15-30 minutes of addition to plasma. Free drug (II) then appears to undergo further metabolism over time. In contrast, free drug (I) is not detected within 15-30 minutes of addition of ADC Molecule 4 to plasma. Levels of free drug (I) increase very slightly over 4 days of incubation in plasma but not in PBS; suggesting a more stable drug linkage in plasma.

EXAMPLE 33

COMPARISON OF ADC CATABOLITES FOR EFFLUX PUMPS

[00615] Permeability glycoprotein 1 (PgP; also known as multidrug resistance protein 1 (MDR1)) is a cell membrane protein that pumps foreign substances out of cells, and reduces intracellular concentrations of a variety of cytotoxic drugs. PgP activity results in blunted chemotherapy-induced cytotoxicity *in vitro* and *in vivo*. Cancer cells frequently become resistant to drugs due to upregulation pf PgP, in some cases this upregulation is mediated by the drug itself. The assay comparing PgP sensitivity of free drugs (I) and (II) (Example 29) conducted in a cisplatin-resistant cell line model.

[00616] To evaluate if free drug (I) is specifically a substrate of P-glycoprotein (PgP), which is responsible for cisplatin-resistance in some of the ovarian cancer cell lines (Stordal *et al.* 2012. *PLoS One* 7(7)), the free drug cell killing activities were investigated on PgP over-expressing MES-SA/MX2 cell line and the parental MES-SA cells. The cell killing activity of free drug (I), free drug (II) and a control free drug (MMAE, designated "III") on the PgP-overexpressing MES-SA/MX2 cells were reduced by different levels compared to their activity on parental MES-SA cells. The MES-SA/MX2 cells were also treated with PgP inhibitor GF120918 (5 μM) to further investigate if the observed cell killing reduction is contributed by the presence of PgP on the cell surface. In the presence of PgP inhibitor, the cell killing activity of free drugs were reversed back to the same level as the parental MES-SA cell line, indicating that PgP overexpression in the MES-SA/MX2 cells were the main reason for the free drug resistance.

[00617] Cell killing EC50 of the positive control free drug (III) on MEA-SA/MX2 cells showed a 111-fold change in the presence and absence of PgP inhibitor GF120918, which indicated that free drug (III) is a very good substrate for PgP. Free drug (I) is a poor substrate for PgP based on the fact that only a 8-fold change in cell killing EC50 was observed in the

presence and absence of PgP inhibitor on MEA-SA/MX2 cells. As a substrate for PgP, free drug (II) is also relatively poor but more susceptible to transport by efflux pumps compared to free drug (I) since a 17-fold change in cell killing EC50 was observed (Table 29, FIG. 33).

Table 29. Cytotoxic activities in Igrov1 and A549 cells

Drug Tested	MEA-SA		MEA-SA/MX2				
	No GF120918		No GF120918		GF120918		EC50 shift
	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	EC50 (nM)	Span (%)	
Free Drug (I)	6	100	50	100	6.4	99	8
Free Drug (II)	1.9	100	28	100	1.7	98	17
Free Drug (III)	0.75	100	111	100	1	99	111

[00618] The data suggests that free drug (I) is a weaker substrate for active transport across the membrane by efflux pumps, compared to free drug (II). As a result, free drug (I) is less likely to be pumped out of cancer cells, which could lead to better cellular retention of the toxin and therefore improved cytotoxicity of ADC Molecule 4 compared to comparator ADC Molecule 21. PgP mediated drug efflux is a common resistance mechanism to ADCs, and in the clinic it is one of the key mechanisms of resistance to platinum agents and PARP inhibitors. The poor substrate capacity of free drug (I) for PgP thus makes it a promising warhead for targeting platinum-resistant and potential PARP-resistant cancers.

EXAMPLE 34

ACCUMULATION OF CATABOLITES IN TUMORS AND PLASMA

[00619] The drug-linkage of ADC Molecule 4 appears to be more stable than that of comparator ADC Molecule 21, however efficient release of free drug (I) from ADC Molecule 4 within the tumor cell is critical for its cytotoxicity. In order to assess warhead release from ADC Molecules 4 and 21, tumor and plasma levels of free drugs (I) and (II) were measured in mice bearing Igrov1 tumors treated with the two ADC molecules. As shown in FIG. 34, release and tumor accumulation of free drug (I) from ADC Molecule 4 was comparable or slightly better than that of free drug (II) from ADC Molecule 21. This data, taken together with the comparable cytotoxicity of the two warheads suggests that the cytotoxic activity of ADC Molecule 4 would be at least comparable to that of comparator ADC Molecule 21.

[00620] To summarize, the data described in Examples 29-32 suggests that a widened therapeutic index (TI) can result from both the attributes of the released warhead (free drug (I) versus free drug (II)) and the architecture of ADC Molecule 4 as a whole. Free drugs (I) and (II) are comparable in *in vitro* cytotoxic activity when administered as free drugs as well as

comparable in their accumulation in tumors when administered as an ADC. The much weaker PgP substrate capability of free drug (I) versus free drug (II) predicts that as tumors develop resistance based on efflux, ADC Molecule 4 will retain most of its original activity. The protease-cleavable release mechanism of ADC Molecule 4 has greater stability, and tumor specificity, than the disulfide release mechanism of comparator ADC Molecule 21. This imparts concomitantly higher specificity for cells expressing FolRα. The greater stability of ADC Molecule 4 together with the faster clearance and higher tolerability of catabolite (I) also confers an improved safety profile for ADC Molecule 4. All of this indicates that ADC Molecule 4 could have a higher TI than comparator ADC IMGN853 as measured on surrogate ADC Molecule 21.

EXAMPLE 35

INTRODUCTION OF MUTATIONS INTO CANDIDATE ANTIBODIES

[00621] A V262E mutation was introduced into antibody variant 1848-H01 (Y180/F404) to investigate whether this mutation would increase yields of the variant. Introduction of the V262E mutation resulted in about a 70% increase in yield post ProA purification (350 mg/L compared to parent titer of 170 mg/L) with no change in quality of purified protein (data not shown). The properties of the V262E mutated protein conjugated to **Conjugate P** (Example 11) and **Conjugate Q** (Example 15) was compared to the parental conjugate ADC Molecule 4 for conjugation efficiency and *in vitro* activity of the ADC. As seen in Table 30, introduction of the V262E mutation reduced the *in vitro* cytotoxic activity on Igrov1 cells of the P conjugate to a small extent and of the Q conjugate to a larger extent, although conjugation efficiency and DAR were comparable.

Table 30. Comparison of ADC molecules with or without V262E mutations

ADC Molecule	Antibody	Conjugate	Conjugation Site(s)	DAR	% Conjugated	Cell Killing (Igrov1)	
						EC50 (nM)	Span (%)
4	1848-H01	P	Y180/F404	3.73	93%	0.083	80
20	1848-H01	Q	Y180/F404	3.81	95%	0.13	70
23	1848-H01 V262E	P	Y180/F404	3.57	89%	0.088	72
24	1848-H01 V262E	Q	Y180/F404	3.76	94%	0.19	56

[00622] A comparison of the pharmacokinetic properties and *in vivo* stability and *in vivo* efficacy of the P conjugate to the ADC Molecule 4 showed that, while PK of the ADCs was comparable between the two versions (Table 31, FIG. 36), the *in vivo* activity of the mutated P

conjugate (ADC Molecule 23) was marginally lower than that of ADC Molecule 4 (FIG. 35A). Statistical analysis of tumor size on day 21 showed that only treatment with ADC Molecule 4 resulted in significantly smaller tumors compared to vehicle, however tumor sizes in the ADC Molecule 4 and ADC Molecule 23 groups were not statistically different from each other (FIG. 35B). Based on this, ADC Molecule 23 is suitable as an alternative ADC for development and further investigation for targeting of FolRα.

Table 31. Pharmacokinetic properties of ADC molecules with or without V262E

ADC Molecule	SP	Antibody	Conjugate	Conjugation Site(s)	Terminal $t_{1/2}$ (hr)	C_0 (μg/mL)	C_{max} ± SE (μg/mL)	$AUC_{0\text{-}last}$ ± SE (day * μg/mL)	$AUC_{0\text{-}inf}$ ± SE (day * μg/mL)	Clearance (mL/days/kg)	Vss (mL/kg)
4	8193	1848-H01	P	Y180/F404	6.36	122	118 ± 5	476 ± 22	523	9.57	79.7
23	8675	1848-H01 V262E	P	Y180/F404	5.70	115	113 ± 11	599 ± 34	636	7.87	60.5

EXAMPLE 36

SEQUENCES

[00623] Table 32 provides sequences referred to herein.

Table 32. Sequences

SEQ ID NO:	Molecule	Region	Scheme	Sequence
1	Human folate receptor alpha (hFOLR1)			MAQRMTTQLLLLLVWVAVVGEAQTRIAW ARTELLNVCMNAKHHKEKPGPEDKLHEQ CRPWRKNACCSTNTSQEAHKDVSYLYRF NWNHCGEMAPACKRHFIQDTCLYECSPN LGPWIQQVDQSWRKERVLNVPLCKEDCE QWWEDCRTSYTCKSNWHKGWNWTSGFNK CAVGAACQPFHFYFPTPTVLCNEIWTHS YKVSNYSRGSGRCIQMWFDPAQGNPNEE VARFYAAAMSGAGPWAAWPFLLSLALML LWLLS
2	Cynomolgus folate receptor alpha			MAQRMTTQLLLLLVWVAVVGEAQTRTAR ARTELLNVCMNAKHHKEKPGPEDKLHEQ CRPWKKNACCSTNTSQEAHKDVSYLYRF NWNHCGEMAPACKRHFIQDTCLYECSPN LGPWIQQVDQSWRKERVLNVPLCKEDCE RWWEDCRTSYTCKSNWHKGWNWTSGFNK CPVGAACQPFHFYFPTPTVLCNEIWTYS YKVSNYSRGSGRCIQMWFDPAQGNPNEE VARFYAAAMSGAGPWAAWPLLLSLALTL LWLLS
3	Murine folate receptor alpha			MAHLMTVQLLLLVMWMAECAQSRATRAR TELLNVCMDAKHHKEKPGPEDNLHDQCS PWKTNSCCSTNTSQEAHKDISYLYRFNW NHCGTMTSECKRHFIQDTCLYECSPNLG PWIQQVDQSWRKERILDVPLCKEDCQQW WEDCQSSFTCKSNWHKGWNWSSGHNECP VGASCHPFTFYFPTSAALCEEIWSHSYK LSNYSRGSGRCIQMWFDPAQGNPNEEVA RFYAEAMSGAGFHGTWPLLCSLSLVLLW VIS
4	SRP1848-A01	CDR-H1	Chothia	GFNITRY
5	SRP1848-A02	CDR-H1	Chothia	GFNISGF
6	SRP1848-A04	CDR-H1	Chothia	GFNIDQS
7	SRP1848-A06	CDR-H1	Chothia	GFNIGNS
8	SRP1848-A07	CDR-H1	Chothia	GFNIGYH
9	SRP1848-A08	CDR-H1	Chothia	GSNIRKH
10	SRP1848-A09	CDR-H1	Chothia	GFNIRKQ
11	SRP1848-A10	CDR-H1	Chothia	GFNIRKY
12	SRP1848-B01	CDR-H1	Chothia	GFNIRNY
13	SRP1848-B03	CDR-H1	Chothia	GFNISMK

SEQ ID NO:	Molecule	Region	Scheme	Sequence
14	SRP1848-B04	CDR-H1	Chothia	SFNISNH
15	SRP1848-B05	CDR-H1	Chothia	GFNISNY
16	SRP1848-B06	CDR-H1	Chothia	GFNISNY
17	SRP1848-B07	CDR-H1	Chothia	GFNISRF
18	SRP1848-B09	CDR-H1	Chothia	GFNITNY
19	SRP1848-B10	CDR-H1	Chothia	GFNTTTK
20	SRP1848-B11	CDR-H1	Chothia	GFNIGNN
21	SRP1848-C01	CDR-H1	Chothia	GFNIGNS
22	SRP1848-C03	CDR-H1	Chothia	GFNIGVY
23	SRP1848-C04	CDR-H1	Chothia	GFNIRHY
24	SRP1848-C05	CDR-H1	Chothia	GFNIRKY
25	SRP1848-C07	CDR-H1	Chothia	GFNIRKY
26	SRP1848-C10	CDR-H1	Chothia	GFNIRTY
27	SRP1848-D02	CDR-H1	Chothia	GFNISHN
28	SRP1848-D03	CDR-H1	Chothia	GFNIRYF
29	SRP1848-D04	CDR-H1	Chothia	GFNISHY
30	SRP1848-D05	CDR-H1	Chothia	GFNISIS
31	SRP1848-D07	CDR-H1	Chothia	GFNISKY
32	SRP1848-D09	CDR-H1	Chothia	GFNISNY
33	SRP1848-D10	CDR-H1	Chothia	GFNISRN
34	SRP1848-E01	CDR-H1	Chothia	GFNITNK
35	SRP1848-E02	CDR-H1	Chothia	GFNIGKY
36	SRP1848-E03	CDR-H1	Chothia	GFNIGNY
37	SRP1848-E05	CDR-H1	Chothia	GFNIGVY
38	SRP1848-E06	CDR-H1	Chothia	GFNINRY
39	SRP1848-E07	CDR-H1	Chothia	GFNIRKS
40	SRP1848-F01	CDR-H1	Chothia	GFNIRTY
41	SRP1848-F02	CDR-H1	Chothia	GFNIRTY
42	SRP1848-F04	CDR-H1	Chothia	GFNISNY
43	SRP1848-F05	CDR-H1	Chothia	GFNISKS
44	SRP1848-F06	CDR-H1	Chothia	GFNISLS
45	SRP1848-F07	CDR-H1	Chothia	GFNISNH
46	SRP1848-F08	CDR-H1	Chothia	GFNISNH
47	SRP1848-F09	CDR-H1	Chothia	GFNISNH
48	SRP1848-F10	CDR-H1	Chothia	GFNISNN
49	SRP1848-F11	CDR-H1	Chothia	GFNISNN
50	SRP1848-G01	CDR-H1	Chothia	GFNISRH
51	SRP1848-G03	CDR-H1	Chothia	GFNISTY
52	SRP1848-G04	CDR-H1	Chothia	GFNIHST
53	SRP1848-G06	CDR-H1	Chothia	GFNIRST
54	SRP1848-G07	CDR-H1	Chothia	GFNIHST
55	SRP1848-G09	CDR-H1	Chothia	GFNIRGT
56	SRP1848-G10	CDR-H1	Chothia	GFNIRST
57	SRP1848-G11	CDR-H1	Chothia	GFNISST

SEQ ID NO:	Molecule	Region	Scheme	Sequence
58	SRP1848-H01	CDR-H1	Chothia	GFNIRTQ
59	SRP2060-E10	CDR-H1	Chothia	GFSLSTFGM
60	SRP2060-E05	CDR-H1	Chothia	GFSLSTFGM
61	SRP2060-B01	CDR-H1	Chothia	GFSLSTFGM
62	SRP2060-A06	CDR-H1	Chothia	GFSLSTFGM
63	SRP1848-A01	CDR-H1	Kabat	RYSIH
64	SRP1848-A02	CDR-H1	Kabat	GFRIH
65	SRP1848-A04	CDR-H1	Kabat	QSSIH
66	SRP1848-A06	CDR-H1	Kabat	NSYIH
67	SRP1848-A07	CDR-H1	Kabat	YHSIH
68	SRP1848-A08	CDR-H1	Kabat	KHSIH
69	SRP1848-A09	CDR-H1	Kabat	KQSIH
70	SRP1848-A10	CDR-H1	Kabat	KYSIH
71	SRP1848-B01	CDR-H1	Kabat	NYSIH
72	SRP1848-B03	CDR-H1	Kabat	MKYIH
73	SRP1848-B04	CDR-H1	Kabat	NHSIH
74	SRP1848-B05	CDR-H1	Kabat	NYYIH
75	SRP1848-B06	CDR-H1	Kabat	NYYIH
76	SRP1848-B07	CDR-H1	Kabat	RFYIH
77	SRP1848-B09	CDR-H1	Kabat	NYYIH
78	SRP1848-B10	CDR-H1	Kabat	TKSIH
79	SRP1848-B11	CDR-H1	Kabat	NNSIH
80	SRP1848-C01	CDR-H1	Kabat	NSYIH
81	SRP1848-C03	CDR-H1	Kabat	VYSIH
82	SRP1848-C04	CDR-H1	Kabat	HYSIH
83	SRP1848-C05	CDR-H1	Kabat	KYSIH
84	SRP1848-C07	CDR-H1	Kabat	KYSIH
85	SRP1848-C10	CDR-H1	Kabat	TYYIH
86	SRP1848-D02	CDR-H1	Kabat	HNYIH
87	SRP1848-D03	CDR-H1	Kabat	YFSIH
88	SRP1848-D04	CDR-H1	Kabat	HYSIH
89	SRP1848-D05	CDR-H1	Kabat	ISYIH
90	SRP1848-D07	CDR-H1	Kabat	KYYIH
91	SRP1848-D09	CDR-H1	Kabat	NYYIH
92	SRP1848-D10	CDR-H1	Kabat	RNSIH
93	SRP1848-E01	CDR-H1	Kabat	NKYIH
94	SRP1848-E02	CDR-H1	Kabat	KYSIH
95	SRP1848-E03	CDR-H1	Kabat	NYYIH
96	SRP1848-E05	CDR-H1	Kabat	VYYIH
97	SRP1848-E06	CDR-H1	Kabat	RYYIH
98	SRP1848-E07	CDR-H1	Kabat	KSSIH
99	SRP1848-F01	CDR-H1	Kabat	TYSIH
100	SRP1848-F02	CDR-H1	Kabat	TYSIH

SEQ ID NO:	Molecule	Region	Scheme	Sequence
101	SRP1848-F04	CDR-H1	Kabat	NYSIH
102	SRP1848-F05	CDR-H1	Kabat	KSSIH
103	SRP1848-F06	CDR-H1	Kabat	LSYIH
104	SRP1848-F07	CDR-H1	Kabat	NHSIH
105	SRP1848-F08	CDR-H1	Kabat	NHSIH
106	SRP1848-F09	CDR-H1	Kabat	NHYIH
107	SRP1848-F10	CDR-H1	Kabat	NNSIH
108	SRP1848-F11	CDR-H1	Kabat	NNYIH
109	SRP1848-G01	CDR-H1	Kabat	RHSIH
110	SRP1848-G03	CDR-H1	Kabat	TYYIH
111	SRP1848-G04	CDR-H1	Kabat	STDIH
112	SRP1848-G06	CDR-H1	Kabat	STDIH
113	SRP1848-G07	CDR-H1	Kabat	STDIH
114	SRP1848-G09	CDR-H1	Kabat	GTDIH
115	SRP1848-G10	CDR-H1	Kabat	STDIH
116	SRP1848-G11	CDR-H1	Kabat	STDIH
117	SRP1848-H01	CDR-H1	Kabat	TQSIH
118	SRP2060-E10	CDR-H1	Kabat	TFGMGVG
119	SRP2060-E05	CDR-H1	Kabat	TFGMGVG
120	SRP2060-B01	CDR-H1	Kabat	TFGMGVG
121	SRP2060-A06	CDR-H1	Kabat	TFGMGVG
122	SRP1848-A01	CDR-H2	Chothia	LPESGG
123	SRP1848-A02	CDR-H2	Chothia	YPESGA
124	SRP1848-A04	CDR-H2	Chothia	YPVDGT
125	SRP1848-A06	CDR-H2	Chothia	TPIDGN
126	SRP1848-A07	CDR-H2	Chothia	FPVDGT
127	SRP1848-A08	CDR-H2	Chothia	YPNDGT
128	SRP1848-A09	CDR-H2	Chothia	FPNDGT
129	SRP1848-A10	CDR-H2	Chothia	FPIDDI
130	SRP1848-B01	CDR-H2	Chothia	YPVDGI
131	SRP1848-B03	CDR-H2	Chothia	TPIDGM
132	SRP1848-B04	CDR-H2	Chothia	YPVDGI
133	SRP1848-B05	CDR-H2	Chothia	SPIDGY
134	SRP1848-B06	CDR-H2	Chothia	TPIDGY
135	SRP1848-B07	CDR-H2	Chothia	SPYDGF
136	SRP1848-B09	CDR-H2	Chothia	TPVDGY
137	SRP1848-B10	CDR-H2	Chothia	YPRDGI
138	SRP1848-B11	CDR-H2	Chothia	SPIDGF
139	SRP1848-C01	CDR-H2	Chothia	TPNDGY
140	SRP1848-C03	CDR-H2	Chothia	YPIDGN
141	SRP1848-C04	CDR-H2	Chothia	YPGPGN
142	SRP1848-C05	CDR-H2	Chothia	FPIDGI
143	SRP1848-C07	CDR-H2	Chothia	FPIDGI

SEQ ID NO:	Molecule	Region	Scheme	Sequence
144	SRP1848-C10	CDR-H2	Chothia	SPIDGY
145	SRP1848-D02	CDR-H2	Chothia	TPQDGY
146	SRP1848-D03	CDR-H2	Chothia	FPNDGS
147	SRP1848-D04	CDR-H2	Chothia	YPRDGI
148	SRP1848-D05	CDR-H2	Chothia	SPIDGY
149	SRP1848-D07	CDR-H2	Chothia	SPNDGY
150	SRP1848-D09	CDR-H2	Chothia	SPNDGY
151	SRP1848-D10	CDR-H2	Chothia	SPNDGT
152	SRP1848-E01	CDR-H2	Chothia	TPFDGF
153	SRP1848-E02	CDR-H2	Chothia	YPNDGN
154	SRP1848-E03	CDR-H2	Chothia	TPRDGF
155	SRP1848-E05	CDR-H2	Chothia	TPNDGY
156	SRP1848-E06	CDR-H2	Chothia	TPNDGY
157	SRP1848-E07	CDR-H2	Chothia	FPYDGS
158	SRP1848-F01	CDR-H2	Chothia	FPNDGT
159	SRP1848-F02	CDR-H2	Chothia	FPNDGT
160	SRP1848-F04	CDR-H2	Chothia	YPIDGI
161	SRP1848-F05	CDR-H2	Chothia	YPNDGS
162	SRP1848-F06	CDR-H2	Chothia	SPIDGN
163	SRP1848-F07	CDR-H2	Chothia	YPNDGI
164	SRP1848-F08	CDR-H2	Chothia	YPVDGI
165	SRP1848-F09	CDR-H2	Chothia	SPLDGY
166	SRP1848-F10	CDR-H2	Chothia	FPNDGY
167	SRP1848-F11	CDR-H2	Chothia	TPIDGN
168	SRP1848-G01	CDR-H2	Chothia	APNDGS
169	SRP1848-G03	CDR-H2	Chothia	TPSDGF
170	SRP1848-G04	CDR-H2	Chothia	TPAGGA
171	SRP1848-G06	CDR-H2	Chothia	TPAGGA
172	SRP1848-G07	CDR-H2	Chothia	TPAGGA
173	SRP1848-G09	CDR-H2	Chothia	TPAGGA
174	SRP1848-G10	CDR-H2	Chothia	TPAGGA
175	SRP1848-G11	CDR-H2	Chothia	TPAGGA
176	SRP1848-H01	CDR-H2	Chothia	FPIDGI
177	SRP2060-E10	CDR-H2	Chothia	WWDDD
178	SRP2060-E05	CDR-H2	Chothia	WWDDD
179	SRP2060-B01	CDR-H2	Chothia	WWDDD
180	SRP2060-A06	CDR-H2	Chothia	WWDDD
181	SRP1848-A01	CDR-H2	Kabat	GILPESGGTSYADSVKG
182	SRP1848-A02	CDR-H2	Kabat	GIYPESGATYYADSVKG
183	SRP1848-A04	CDR-H2	Kabat	VIYPVDGTTDYADSVKG
184	SRP1848-A06	CDR-H2	Kabat	GITPIDGNTDYADSVKG
185	SRP1848-A07	CDR-H2	Kabat	EIFPVDGTTDYADSVKG
186	SRP1848-A08	CDR-H2	Kabat	SIYPNDGTTDYADSVKG

SEQ ID NO:	Molecule	Region	Scheme	Sequence
187	SRP1848-A09	CDR-H2	Kabat	SIFPNDGTTDYADSVKG
188	SRP1848-A10	CDR-H2	Kabat	DIFPIDDITDYADSVKG
189	SRP1848-B01	CDR-H2	Kabat	EIYPVDGITDYADSVKG
190	SRP1848-B03	CDR-H2	Kabat	GITPIDGMTDYADSVKG
191	SRP1848-B04	CDR-H2	Kabat	EIYPVDGITDYADSVKG
192	SRP1848-B05	CDR-H2	Kabat	GISPIDGYTDYADSMKG
193	SRP1848-B06	CDR-H2	Kabat	GITPIDGYTDYADSVKG
194	SRP1848-B07	CDR-H2	Kabat	GISPYDGFTDYADSVKG
195	SRP1848-B09	CDR-H2	Kabat	GITPVDGYTDYADRVKG
196	SRP1848-B10	CDR-H2	Kabat	EIYPRDGITDYADSVKG
197	SRP1848-B11	CDR-H2	Kabat	DISPIDGFTDYADSVKG
198	SRP1848-C01	CDR-H2	Kabat	GVTPNDGYTDYADSVKG
199	SRP1848-C03	CDR-H2	Kabat	EIYPIDGNTDYADSVKG
200	SRP1848-C04	CDR-H2	Kabat	EIYPGPGNTDYADSVKG
201	SRP1848-C05	CDR-H2	Kabat	DIFPIDGINDYADSVKG
202	SRP1848-C07	CDR-H2	Kabat	DIFPIDGITDYADSVKG
203	SRP1848-C10	CDR-H2	Kabat	GISPIDGYTDYADSMKG
204	SRP1848-D02	CDR-H2	Kabat	GITPQDGYTDYADSVKG
205	SRP1848-D03	CDR-H2	Kabat	DIFPNDGSTDYADSVKG
206	SRP1848-D04	CDR-H2	Kabat	EIYPRDGITDYADSVKG
207	SRP1848-D05	CDR-H2	Kabat	GISPIDGYTDYADSVKG
208	SRP1848-D07	CDR-H2	Kabat	GISPNDGYTDYADSVKG
209	SRP1848-D09	CDR-H2	Kabat	GISPNDGYTDYADSVKG
210	SRP1848-D10	CDR-H2	Kabat	WISPNDGTTDYADSVKG
211	SRP1848-E01	CDR-H2	Kabat	GITPFDGFTDYADSVKG
212	SRP1848-E02	CDR-H2	Kabat	EIYPNDGNTDYADSVKG
213	SRP1848-E03	CDR-H2	Kabat	GITPRDGFTDYADSVKG
214	SRP1848-E05	CDR-H2	Kabat	GITPNDGYTDYADSVKG
215	SRP1848-E06	CDR-H2	Kabat	GITPNDGYTDYADSVEG
216	SRP1848-E07	CDR-H2	Kabat	EIFPYDGSTDYADNVKG
217	SRP1848-F01	CDR-H2	Kabat	SIFPNDGTTDYADSVKG
218	SRP1848-F02	CDR-H2	Kabat	SIFPNDGTTDYADSVKG
219	SRP1848-F04	CDR-H2	Kabat	EIYPIDGITDYADSVKG
220	SRP1848-F05	CDR-H2	Kabat	EIYPNDGSTDYADSVKG
221	SRP1848-F06	CDR-H2	Kabat	GISPIDGNTDYADSVKG
222	SRP1848-F07	CDR-H2	Kabat	EIYPNDGITDYADSVKG
223	SRP1848-F08	CDR-H2	Kabat	EIYPVDGITDYADSVKG
224	SRP1848-F09	CDR-H2	Kabat	GISPLDGYTDYADSVKG
225	SRP1848-F10	CDR-H2	Kabat	SIFPNDGYTDYADSVKG
226	SRP1848-F11	CDR-H2	Kabat	GITPIDGNTDYADSVKG
227	SRP1848-G01	CDR-H2	Kabat	WIAPNDGSTDYADSVKG
228	SRP1848-G03	CDR-H2	Kabat	GITPSDGFTDYADSVKG
229	SRP1848-G04	CDR-H2	Kabat	YITPAGGATFYADSVKG
230	SRP1848-G06	CDR-H2	Kabat	YITPAGGATYYADNVKG

SEQ ID NO:	Molecule	Region	Scheme	Sequence
231	SRP1848-G07	CDR-H2	Kabat	YITPAGGATWYADSVKG
232	SRP1848-G09	CDR-H2	Kabat	YITPAGGATFYADSVKG
233	SRP1848-G10	CDR-H2	Kabat	YITPAGGATYYADSVKG
234	SRP1848-G11	CDR-H2	Kabat	YITPAGGATWYADSVKG
235	SRP1848-H01	CDR-H2	Kabat	DIFPIDGITDYADSVKG
236	SRP2060-E10	CDR-H2	Kabat	HIWWDDDKYYHPALKG
237	SRP2060-E05	CDR-H2	Kabat	HIWWDDDKYYHPALKG
238	SRP2060-B01	CDR-H2	Kabat	HIWWDDDKYYHPALKG
239	SRP2060-A06	CDR-H2	Kabat	HIWWDDDKYYYPALKG
240	SRP1848-A01	CDR-H3		HIYPWDWFSNYVLDY
241	SRP1848-A02	CDR-H3		HLYVWDWVLDHVLDY
242	SRP1848-A04	CDR-H3		GAWSWRSGYGYYIDY
243	SRP1848-A06	CDR-H3		GAWSWRSGYGYYIDY
244	SRP1848-A07	CDR-H3		GFWAWRSGYGYYLDY
245	SRP1848-A08	CDR-H3		GSWFWRAGYGYYLDY
246	SRP1848-A09	CDR-H3		GSWFWRSGYGYFLEY
247	SRP1848-A10	CDR-H3		GSWSWPSGHSYYLDY
248	SRP1848-B01	CDR-H3		GFWSWPSGYSYFLDY
249	SRP1848-B03	CDR-H3		GSWSWPSGYSYYLDY
250	SRP1848-B04	CDR-H3		GRYSWRAGYSYYLDY
251	SRP1848-B05	CDR-H3		GSWFWQSGYGYYLDY
252	SRP1848-B06	CDR-H3		GFWSWPSGYGYYQDY
253	SRP1848-B07	CDR-H3		GSWSWPAGYGYYQDY
254	SRP1848-B09	CDR-H3		GAWSWRSGYGYYMDY
255	SRP1848-B10	CDR-H3		GGWHWRSGYSYYLDY
256	SRP1848-B11	CDR-H3		GSWSWRAGYGYYLDY
257	SRP1848-C01	CDR-H3		GSWFWRAGYGYYLDY
258	SRP1848-C03	CDR-H3		GSWAWRSGYSYYLDY
259	SRP1848-C04	CDR-H3		GSLSWRAGYGYYLDY
260	SRP1848-C05	CDR-H3		GSWSWKAGYGYYLDY
261	SRP1848-C07	CDR-H3		GSWSWPAGYGYYQDY
262	SRP1848-C10	CDR-H3		GSWSWPAGYGYYLDY
263	SRP1848-D02	CDR-H3		GAWSWRAGYGYYLDY
264	SRP1848-D03	CDR-H3		GHWSWPSGYWYYLDY
265	SRP1848-D04	CDR-H3		GYWFWRSGYGYYLDY
266	SRP1848-D05	CDR-H3		GSWSWRAGYGYYLDY
267	SRP1848-D07	CDR-H3		GFWAWRSGYGYYLDY
268	SRP1848-D09	CDR-H3		GSWSWRHGYGYYLDY
269	SRP1848-D10	CDR-H3		GAWSWRSGYGYYIDY
270	SRP1848-E01	CDR-H3		GSWSWPAGYGYYQDY
271	SRP1848-E02	CDR-H3		GSWSWRSGYGYYLDY
272	SRP1848-E03	CDR-H3		GSWSWPAGHSYYLDY
273	SRP1848-E05	CDR-H3		GFWAWRSGYGYYLDY

SEQ ID NO:	Molecule	Region	Scheme	Sequence
274	SRP1848-E06	CDR-H3		GTWSWPSGHSYYLDY
275	SRP1848-E07	CDR-H3		GAWSWRSGYGYYIDY
276	SRP1848-F01	CDR-H3		GSWAWRAGYSYYLDY
277	SRP1848-F02	CDR-H3		GSWSWQAGYGYYLDY
278	SRP1848-F04	CDR-H3		GSWFWRSGYGYYLDY
279	SRP1848-F05	CDR-H3		GSWAWRSGYSYFLDY
280	SRP1848-F06	CDR-H3		GFWAWRSGYGYYLDY
281	SRP1848-F07	CDR-H3		GSWDWRSGYSYYLDY
282	SRP1848-F08	CDR-H3		GSWYWQSGYSYYLDY
283	SRP1848-F09	CDR-H3		GAWSWRSGYGYYIDY
284	SRP1848-F10	CDR-H3		GSWFWRSGYGYYLDY
285	SRP1848-F11	CDR-H3		GSWYWRAGYGYYLDY
286	SRP1848-G01	CDR-H3		GSWAWRSGYSYFLDY
287	SRP1848-G03	CDR-H3		GSWSWPSGHGYFLDY
288	SRP1848-G04	CDR-H3		YPYWFAGYMDY
289	SRP1848-G06	CDR-H3		QPYWFAGYMDY
290	SRP1848-G07	CDR-H3		YPFWFAGYMDY
291	SRP1848-G09	CDR-H3		HEYWFSGYMDY
292	SRP1848-G10	CDR-H3		YPYWFAGYIDY
293	SRP1848-G11	CDR-H3		YPYWFSGYMDY
294	SRP1848-H01	CDR-H3		GSWSWPSGMDYYLDY
295	SRP2060-E10	CDR-H3		NHFPHYYGSSHWYFNV
296	SRP2060-E05	CDR-H3		NHFPHYYGSSHWYFNV
297	SRP2060-B01	CDR-H3		NHFPHYYGSSHWYFNV
298	SRP2060-A06	CDR-H3		NHFPHYYGSSHWYFDV
299	trastuzumab	CDR-L1		RASQDVNTAVA
300	H6D1-LC4	CDR-L1		KASQDINSYLS
301	H6D1-LC5	CDR-L1		KASQDINSYLS
302	trastuzumab	CDR-L2		SASFLYS
303	H6D1-LC4	CDR-L3		RANRLVD
304	H6D1-LC5	CDR-L2		RANRLVD
305	trastuzumab	CDR-L3		QQHYTTPPT
306	H6D1-LC4	CDR-L3		LQYDEFPYT
307	H6D1-LC5	CDR-L3		LQYDEFPYT
308	SRP1848-A01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ITRYSIHWVRQAPGKGLEWVAGILPESG GTSYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARHIYPWDWFSNYVLD YWGQGTLVTVSS
309	SRP1848-A02	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISGFRIHWVRQAPGKGLEWVAGIYPESG ATYYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARHLYVWDWVLDHVLD YWGQGTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
310	SRP1848-A04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IDQSSIHWVRQAPGKGLEWVGVIYPVDG TTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYID YWGQGTLVTVSS
311	SRP1848-A06	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGNSYIHWVRQAPGKGLEWVGGITPIDG NTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYID YWGQGTLVTVSS
312	SRP1848-A07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGYHSIHWVRQAPGKGLEWVGEIFPVDG TTDYADSVKGRFTISADTSKNTAYLHMN SLRAEDTAVYYCARGFWAWRSGYGYYLD YWGQGTLVTVSS
313	SRP1848-A08	VH		EVQLVESGGGLVQPGGSLRLSCAASGSN IRKHSIHWVRQAPGKGLEWVGSIYPNDG TTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWFWRAGYGYYLD YWGQGTLVTVSS
314	SRP1848-A09	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRKQSIHWVRQAPGKGLEWVGSIFPNDG TTDYADSVKGRFTISADTSKNTAYLQVN SLRAEDTAVYYCARGSWFWRSGYGYFLE YWGQGTLVTVSS
315	SRP1848-A10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRKYSIHWARQAPGKGLEWVGDIFPIDD ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPSGHSYYLD YWGQGTLVTVSS
316	SRP1848-B01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRNYSIHWVRQAPGKGLEWVGEIYPVDG ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGFWSWPSGYSYFLD YWGQGTLVTVSS
317	SRP1848-B03	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISMKYIHWVRQAPGKGLEWVGGITPIDG MTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPSGYSYYLD YWGQGTLVTVSS
318	SRP1848-B04	VH		EVQLVESGGGLVQPGGSLRLSCAASSFN ISNHSIHWVRQAPGKGLEWVGEIYPVDG ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGRYSWRAGYSYYLD YWGQGTLVTVSS
319	SRP1848-B05	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNYYIHWVRQAPGKGLEWVGGISPIDG YTDYADSMKGRFTISADTSKNTAYLQMS SLRAEDTAVYYCARGSWFWQSGYGYYLD YWGQGTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
320	SRP1848-B06	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNYYIHWVRQAPGKGLEWVGGITPIDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGFWSWPSGYGYYQD YWGQGTLVTVSS
321	SRP1848-B07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISRFYIHWVRQAPGKGLEWVGGISPYDG FTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPAGYGYYQD YWGQGTLVTVSS
322	SRP1848-B09	VH		EVQLVESGGGLVQPGGSLRLSCAAGGFN ITNYYIHWVRQAPGKGLEWVGGITPVDG YTDYADRVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYMD YWGQGTLVTVSS
323	SRP1848-B10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN TTTKSIHWVRQAPGKGLEWVGEIYPRDG ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGGWHWRSGYSYYLD YWGQGTLVTVSS
324	SRP1848-B11	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGNNSIHWVRQAPGKGLEWVGDISPIDG FTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWRAGYGYYLD YWGQGTLVTVSS
325	SRP1848-C01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGNSYIHWVRQAPGKGLEWVGGVTPNDG YTDYADSVKGRFTISADTSKNTTYLQMN SLRAEDTAVYYCARGSWFWRAGYGYYLD YWGQGALVTVSS
326	SRP1848-C03	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGVYSIHWVRQAPGKGLEWVGEIYPIDG NTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWAWRSGYSYYLD YWGQGTLVTVSS
327	SRP1848-C04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRHYSIHWVRQAPGKGLEWVGEIYPGPG NTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSLSWRAGYGYYLD YWGQGTLVTVSS
328	SRP1848-C05	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRKYSIHWVRQAPGKGLEWVGDIFPIDG INDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWKAGYGYYLD YWGQGTLVTVSS
329	SRP1848-C07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRKYSIHWVRQAPGKGLEWVGDIFPIDG ITDYADSMKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPAGYGYYQD YWGQGTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
330	SRP1848-C10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRTYYIHWVRQAPGKGLEWVGGISPIDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPAGYGYYLD YWGQGTLVTVSS
331	SRP1848-D02	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISHNYIHWVRQAPGKGLEWVGGITPQDG YTDYADSVKGRFTISADTSKNTAYLQMN RLRAEDTAVYYCARGAWSWRAGYGYYLD YWGQGTLVTVSS
332	SRP1848-D03	VH		EVQLVESGGGVVQPGGSLRLSCAASGFN IRYFSIHWVRQAPGKGLEWVGDIFPNDG STDYADSVKGRFTISADTSKNTAYLQMN SLRAEETAVYYCARGHWSWPSGYWYYLD YWGQGTLVTVSS
333	SRP1848-D04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISHYSIHWVRQAPGKGLEWVGEIYPRDG ITDYADSVKGRFTISADTSKNTAYLQMN SLSAEDTAVYYCARGYWFWRSGYGYYLD YWGQGTLVTVSS
334	SRP1848-D05	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISISYIHWVRQAPGKGLEWVGGISPIDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWRAGYGYYLD YWGQGTLVTVSS
335	SRP1848-D07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISKYYIHWVRQAPGKGLEWVGGISPNDG YTDYADSVKGRFAISADTSKNTAYLQMN SLRAEDTAVYYCARGFWAWRSGYGYYLD YWGQGTLVTVSS
336	SRP1848-D09	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNYYIHWVRQAPGKGLEWVGGISPNDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWRHGYGYYLD YWGQGTLVTVSS
337	SRP1848-D10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISRNSIHWVRQAPGKGLEWVGWISPNDG TTDYADSVKGRFTISADGSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYID YWGQGTLVTVSS
338	SRP1848-E01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ITNKYIHWVRQAPGKGLEWVGGITPFDG FTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPAGYGYYQD YWGQGTLVTVSS
339	SRP1848-E02	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IGKYSIHWVRQAPGKGLEWVGEIYPNDG NTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWRSGYGYYLD YWGQGTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
340	SRP1848-E03	VH		EVQLVESGGGLAQPGGSLRLSCAASGFN IGNYYIHWVRQAPGKGLEWVGGITPRDG FTDYADSVKGRFTISADTSKNTAYLQVN SLRAEDTAVYYCARGSWSWPAGHSYYLD YWGQGTLVTVSS
341	SRP1848-E05	VH		EVQLVESGGGLVQPGGSLRVSCAASGFN IGVYYIHWVRQAPGKGLEWVGGITPNDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGFWAWRSGYGYYLD YWGQGTLVTVSS
342	SRP1848-E06	VH		EVQLVESGGGLVQPSGSLRLSCAASGFN INRYYIHWVRQAPGKGLEWVGGITPNDG YTDYADSVEGRFTTSADTSKNTAYLQMN SLRAEDTAVYYCARGTWSWPSGHSYYLD YWGQGTLVTVSS
343	SRP1848-E07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRKSSIHWVRQAPGKGLEWVGEIFPYDG STDYADNVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYID YWGQGTLVTVSS
344	SRP1848-F01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRTYSIHWVRQAPGKGLEWVGSIFPNDG TTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWAWRAGYSYYLD YWGQGTLVTVSS
345	SRP1848-F02	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRTYSIHWVRQAPGKGLEWVGSIFPNDG TTDYADSVKGRLTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWQAGYGYYLD YWGQGTLVTVSS
346	SRP1848-F04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNYSIHWVRQAPGKGLEWVGEIYPIDG ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWFWRSGYGYYLD YWGQGTLVTVSS
347	SRP1848-F05	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISKSSIHWVRQAPGKGLEWVGEIYPNDG STDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWAWRSGYSYFLD YWGQGTLVTVSS
348	SRP1848-F06	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISLSYIHWVRQAPGKGLEWVGGISPIDG NTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGFWAWRSGYGYYLD YWGQGTLVTVSS
349	SRP1848-F07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNHSIHWVRQAPGKGLEWVGEIYPNDG ITDYADSVKGRFTISADTSKNTAYLQMN SLSAEDTAVYYCARGSWDWRSGYSYYLD YWGQGTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
350	SRP1848-F08	VH		EVQLVESGGGLVQPGGSLRLSCAAGGFN ISNHSIHWVRQAPGKGVEWVGEIYPVDG ITDYADSVKGRFTISADTSKNTAYLRMN SLRAEDTAVYYCARGSWYWQSGYSYYLD YWGQGTLVTVSS
351	SRP1848-F09	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNHYIHWVRQAPGKGLEWVGGISPLDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGAWSWRSGYGYYID YWGQGTLVTVSS
352	SRP1848-F10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNNSIHWVRQAPGKGLEWVGSIFPNDG YTDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWFWRSGYGYYLD YWGQGTLVTVSS
353	SRP1848-F11	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISNNYIHWVRQAPGKGLEWVGGITPIDG NTDYADSVKGRFTISADTSMNTAYLQMN SLRAEDTAVYYCARGSWYWRAGYGYYLD YWGQGALVTVSS
354	SRP1848-G01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISRHSIHWVRQAPGKGLEWVGWIAPNDG STDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWAWRSGYSYFLD YWGQGTLVTVSS
355	SRP1848-G03	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISTYYIHWVRQAPGKGLEWVGGITPSDG FTDYADSVKGRSTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPSGHGYFLD YWGQGTLVTVSS
356	SRP1848-G04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IHSTDIHWVRQAPGKGLEWVAYITPAGG ATFYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARYPYWFAGYMDYWGQ GTLVTVSS
357	SRP1848-G06	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRSTDIHWVRQAPGKGLEWVAYITPAGG ATYYADNVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARQPYWFAGYMDYWGQ GTLVTVSS
358	SRP1848-G07	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IHSTDIHWVRQAPGKGLEWVAYITPAGG ATWYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARYPFWFAGYMDYWGQ GTLVTVSS
359	SRP1848-G09	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRGTDIHWVRQAPGKGLEWVAYITPAGG ATFYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARHEYWFSGYMDYWGQ GTLVTVSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
360	SRP1848-G10	VH		EVQLVESGGGLVQPGSSLRLSCAASGFN IRSTDIHWVRQAPGKGLEWVAYITPAGG ATYYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARYPYWFAGYIDYWGQ GTLVTVSS
361	SRP1848-G11	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN ISSTDIHWVRQAPGKGLEWVAYITPAGG ATWYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARYPYWFSGYMDYWGQ GTLVTVSS
362	SRP1848-H01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFN IRTQSIHWVRQAPGKGLEWIGDIFPIDG ITDYADSVKGRFTISADTSKNTAYLQMN SLRAEDTAVYYCARGSWSWPSGMDYYLD YWGQGTLVTVSS
363	SRP2060-E10	VH		EVQLLESGGGLVQPGGSLRLSCAFSGFS LSTFGMGVGWVRQAPGKGLEWVSHIWWD DDKYYHPALKGRFTISKDNSKNTVYLQM NSLRAEDTAVYYCGRNHFPHYYGSSHWY FNVWGQGTTVTVSS
364	SRP2060-E05	VH		EVQLLESGGGLVQPGGSLRLSCAFSGFS LSTFGMGVGWVRQAPGKGLEWVSHIWWD DDKYYHPALKGRFTVSKDNSKNTVYLQM NSLRAEDTAVYYCGRNHFPHYYGSSHWY FNVWGQGTTVTVSS
365	SRP2060-B01	VH		EVQLLESGGGLVQPGGSLRLSCALSGFS LSTFGMGVGWVRQATGKGLEWVSHIWWD DDKYYHPALKGRFTISKDNSKNTVHLQM NSLRAEDTAVYYCGRNHFPHYYGSSHWY FNVWGQGTTVTVSS
366	SRP2060-A06	VH		EVQLLESGGGLVQPGGSLRLSCAFSGFS LSTFGMGVGWVRQAPGKGLEWVGHIWWD DDKYYYPALKGRFTISKDNSKNTVYLQM NSLRAEDTAVYYCGRNHFPHYYGSSHWY FDVWGQGTTVTVSS
367	trastuzumab	VL		DIQMTQSPSSLSASVGDRVTITCRASQD VNTAVAWYQQKPGKAPKLLIYSASFLYS GVPSRFSGSRSGTDFTLTISSLQPEDFA TYYCQQHYTTPPTFGQGTKVEIK
368	H6D1-LC4	VL		EIVMTQSPATLSLSPGERATLSCKASQD INSYLSWYQQKPGQAPRLLIYRANRLVD GIPARFSGSGSGTDYTLTISSLEPEDFA VYYCLQYDEFPYTFGGGTKVEIK
369	H6D1-LC5	VL		DIQMTQSPSTLSASVGDRVTITCKASQD INSYLSWYQQKPGKAPKLLIYRANRLVD GVPSRFSGSGSGTEFTLTISSLQPDDFA TYYCLQYDEFPYTFGGGTKVEIK

SEQ ID NO:	Molecule	Region	Scheme	Sequence
370	Human IgG1 HC Constant			ASTKGPSVFPLAPSSKSTSGGTAALGCL VKDYFPEPVTVSWNSGALTSGVHTFPAV LQSSGLYSLSSVVTVPSSSLGTQTYICN VNHKPSNTKVDKKVEPKSCDKTHTCPPC PAPELLGGPSVFLFPPKPKDTLMISRTP EVTCVVVDVSHEDPEVKFNWYVDGVEVH NAKTKPREEQYNSTYRVVSVLTVLHQDW LNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLV KGFYPSDIAVEWESNGQPENNYKTTPPV LDSDGSFFLYSKLTVDKSRWQQGNVFSC SVMHEALHNHYTQKSLSLSPGK
371	Human IgG LC Constant Ckappa			RTVAAPSVFIFPPSDEQLKSGTASVVCL LNNFYPREAKVQWKVDNALQSGNSQESV TEQDSKDSTYSLSSTLTLSKADYEKHKV YACEVTHQGLSSPVTKSFNRGEC
372	Mouse IgG1 HC Constant			AKTTPPSVYPLAPGSAAQTNSMVTLGCL VKGYFPEPVTVTWNSGSLSSGVHTFPAV LQSDLYTLSSSVTVPSSTWPSETVTCNV AHPASSTKVDKKIVPRDCGCKPCICTVP EVSSVFIFPPKPKDVLTITLTPKVTCVV VDISKDDPEVQFSWFVDDVEVHTAQTQP REEQFNSTFRSVSELPIMHQDWLNGKEF KCRVNSAAFPAPIEKTISKTKGRPKAPQ VYTIPPPKEQMAKDKVSLTCMITDFFPE DITVEWQWNGQPAENYKNTQPIMDTDGS YFVYSKLNVQKSNWEAGNTFTCSVLHEG LHNHHTEKSLSHSPG
373	Mouse IgG LC Constant Ckappa			RADAAPTVSIFPPSSEQLTSGGASVVCF LNNFYPKDINVKWKIDGSERQNGVLNSW TDQDSKDSTYSMSSTLTLTKDEYERHNS YTCEATHKTSTSPIVKSFNRNEC
374	Kappa LC			HMTVAAPSVFIFPPSDEQLKSGTASVVC LLNNFYPREAKVQWKVDNALQSGNSQES VTEQDSKDSTYSLSSTLTLSKADYEKHK VYACEVTHQGLSSPVTKSFNRGEC
375	Lambda LD			GQPKAAPSVTLFPPSSEELQANKATLVC LISDFYPGAVTVAWKADSSPVKAGVETT TPSKQSNNKYAASSYLSLTPEQWKSHRS YSCQVTHEGSTVEKTVAPTECS
376	FlagHis Tag			GSGDYKDDDDKGSGHHHHHH
377	Linker			GGGGSGGGGSGGGGS
378	Linker			AAGSDQEPKSS

SEQ ID NO:	Molecule	Region	Scheme	Sequence
379	1848-B10-VH-(G4S)3-VL	scFv		MEVQLVESGGGLVQPGGSLRLSCAASGF NTTTKSIHWVRQAPGKGLEWVGEIYPRD GITDYADSVKGRFTISADTSKNTAYLQM NSLRAEDTAVYYCARGGWHWRSGYSYYL DYWGQGTLVTVSSGGGGSGGGGSGGGGS DIQMTQSPSSLSASVGDRVTITCRASQD VNTAVAWYQQKPGKAPKLLIYSASFLYS GVPSRFSGSRSGTDFTLTISSLQPEDFA TYYCQQHYTTPPTFGQGTKVEIK
380	1848-B10-VL-(G4S)3-VH	scFv		MDIQMTQSPSSLSASVGDRVTITCRASQ DVNTAVAWYQQKPGKAPKLLIYSASFLY SGVPSRFSGSRSGTDFTLTISSLQPEDF ATYYCQQHYTTPPTFGQGTKVEIKGGGG SGGGGSGGGGSEVQLVESGGGLVQPGGS LRLSCAASGFNTTTKSIHWVRQAPGKGL EWVGEIYPRDGITDYADSVKGRFTISAD TSKNTAYLQMNSLRAEDTAVYYCARGGW HWRSGYSYYLDYWGQGTLVTVSS
381	1848-B10-VH-(G4S)3-VL	scFv-Fc		MEVQLVESGGGLVQPGGSLRLSCAASGF NTTTKSIHWVRQAPGKGLEWVGEIYPRD GITDYADSVKGRFTISADTSKNTAYLQM NSLRAEDTAVYYCARGGWHWRSGYSYYL DYWGQGTLVTVSSGGGGSGGGGSGGGGS DIQMTQSPSSLSASVGDRVTITCRASQD VNTAVAWYQQKPGKAPKLLIYSASFLYS GVPSRFSGSRSGTDFTLTISSLQPEDFA TYYCQQHYTTPPTFGQGTKVEIKAAGSD QEPKSSDKTHTCPPCPAPELLGGPSVFL FPPKPKDTLMISRTPEVTCVVVDVSHED PEVKFNWYVDGVEVHNAKTKPREEQYNS TYRVVSVLTVLHQDWLNGKEYKCKVSNK ALPAPIEKTISKAKGQPREPQVYTLPPS REEMTKNQVSLTCLVKGFYPSDIAVEWE SNGQPENNYKTTPPVLDSDGSFFLYSKL TVDKSRWQQGNVFSCSVMHEALHNHYTQ KSLSLSPGK

SEQ ID NO:	Molecule	Region	Scheme	Sequence
382	1848-B10-VL-(G4S)3-VH	scFv-Fc		MDIQMTQSPSSLSASVGDRVTITCRASQ DVNTAVAWYQQKPGKAPKLLIYSASFLY SGVPSRFSGSRSGTDFTLTISSLQPEDF ATYYCQQHYTTPPTFGQGTKVEIKGGGG SGGGGSGGGGSEVQLVESGGGLVQPGGS LRLSCAASGFNTTTKSIHWVRQAPGKGL EWVGEIYPRDGITDYADSVKGRFTISAD TSKNTAYLQMNSLRAEDTAVYYCARGGW HWRSGYSYYLDYWGQGTLVTVSSAAGSD QEPKSSDKTHTCPPCPAPELLGGPSVFL FPPKPKDTLMISRTPEVTCVVVDVSHED PEVKFNWYVDGVEVHNAKTKPREEQYNS TYRVVSVLTVLHQDWLNGKEYKCKVSNK ALPAPIEKTISKAKGQPREPQVYTLPPS REEMTKNQVSLTCLVKGFYPSDIAVEWE SNGQPENNYKTTPPVLDSDGSFFLYSKL TVDKSRWQQGNVFSCSVMHEALHNHYTQ KSLSLSPGK

Equivalents

[00624] The disclosure set forth above may encompass multiple distinct inventions with independent utility. Although each of these inventions has been disclosed in its preferred form(s), the specific embodiments thereof as disclosed and illustrated herein are not to be considered in a limiting sense, because numerous variations are possible. The subject matter of the inventions includes all novel and nonobvious combinations and subcombinations of the various elements, features, functions, and/or properties disclosed herein. The following claims particularly point out certain combinations and subcombinations regarded as novel and nonobvious. Inventions embodied in other combinations and subcombinations of features, functions, elements, and/or properties may be claimed in this application, in applications claiming priority from this application, or in related applications. Such claims, whether directed to a different invention or to the same invention, and whether broader, narrower, equal, or different in scope in comparison to the original claims, also are regarded as included within the subject matter of the inventions of the present disclosure.

[00625] One or more features from any embodiments described herein or in the figures may be combined with one or more features of any other embodiments described herein or in the figures without departing from the scope of the invention.

[00626] All publications, patents and patent applications cited in this specification are herein incorporated by reference as if each individual publication or patent application were specifically and individually indicated to be incorporated by reference. Although the foregoing invention has been described in some detail by way of illustration and example for purposes of clarity of understanding, it will be readily apparent to those of ordinary skill in the art in light of the teachings of this invention that certain changes and modifications may be made thereto without departing from the spirit or scope of the appended claims.

WHAT IS CLAIMED IS:

1. A composition comprising:

(a) an antibody conjugate comprising an antibody that specifically binds to folate receptor alpha (FOLR1) linked site-specifically to at least one payload moiety, wherein the antibody comprises one or more non-natural amino acids at sites selected from the group consisting of: HC-F404, HC-K121, HC-Y180, HC-F241, HC-221, LC-T22, LC-S7, LC-N152, LC-K42, LC-E161, LC-D170, HC-S136, HC-S25, HC-A40, HC-S119, HC-S190, HC-K222, HC-R19, HC-Y52, or HC-S70, according to the Kabat, Chothia, or EU numbering scheme; and

(b) one or more PD-1 or PD-L1 inhibitors.

2. The composition of claim 1, wherein the one or more PD-1 or PD-L1 inhibitors comprise a small molecule blocker of the PD-1 or PD-L1 pathway.

3. The composition of claim 1, wherein the one or more PD-1 or PD-L1 inhibitors comprise an antibody that inhibits PD-1 or PD-L1 activity.

4. The composition of any one of claims 1 to 3, wherein the one or more PD-1 or PD-L1 inhibitors are selected from the group consisting of: CA-170, BMS-8, BMS-202, BMS-936558, CK-301, AUNP12, avelumab, nivolumab, pembrolizumab, atezolizumab, durvalumab, AMP-224, MEDI0680/AMP-514, PDR001, cemiplimab, TSR-042, Tizlelizumab/BGB-A317, CK-301, BMS-936559, camrelizumab, sintilimab, toripalimab, genolimzumab, A167, MGA012, PF-06801591, LY3300054, FAZ053, PD-11, CX-072, BGB-A333, BI 754091, JNJ-63723283, AGEN2034, CA-327, CX-188, STI –A1110, JTX-4014, (LLY) AM0001, CBT-502, FS118, XmAb20717, XmAb23104, AB122, KY1003, RXI-762, PRS-33, ALPN-202, TSR-075, MCLA-145, MGD013, and MGD019.

5. The composition of claim 4, wherein the one or more PD-1 or PD-L1 inhibitors comprise avelumab.

6. The composition of claim 4, wherein the one or more PD-1 or PD-L1 inhibitors comprise pembrolizumab.

7. The composition of claim 3, wherein the antibody that inhibits PD-1 or PD-L1 activity is selected from the antibodies disclosed in WO 2016/077397, WO 2018/156777, and International Application No. PCT/US2013/034213, filed May 23, 2018.

8. The composition of any one of claims 1 to 7, wherein the one or more non-natural amino acids is selected from the group consisting of p-acetyl-L-phenylalanine, O-methyl-L-tyrosine, an -3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, a tri-O-acetyl-GlcNAcβ-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-azido-L-phenylalanine, p-azido-methyl-L-phenylalanine, compound 56, p-acyl-L-phenylalanine, p-benzoyl-L-phenylalanine, L-phosphoserine, phosphonoserine, phosphonotyrosine, p-iodo-phenylalanine, p-bromophenylalanine, p-amino-L-phenylalanine, isopropyl-L-phenylalanine, and p-propargyloxy-phenylalanine.

9. The composition of claim 8, wherein a residue of the one or more non-natural amino acids is linked to the payload moiety via a linker that is hydrolytically stable.

10. The composition of claim 8, wherein a residue of the one or more non-natural amino acids is linked to the payload moiety via a linker that is cleavable.

11. The composition of any one of claims 8 to 10, wherein the non-natural amino acid residue is a residue of compound (30) or compound (56).

12. The composition of any one of the preceding claims wherein the payload moiety is selected from the group consisting of maytansines, hemiasterlins, amanitins, and auristatins.

13. The composition of any one of the preceding claims wherein the payload moiety is selected from the group consisting of DM1, hemiasterlin, amanitin, MMAF, and MMAE.

14. The composition of any one of the preceding claims, wherein the antibody comprises:

(i) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 58 and 117; a CDR-H2 comprising one of SEQ ID NOs: 176 and 235; and a CDR-H3 comprising SEQ ID NO: 294;

(ii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 19 and 78; a CDR-H2 comprising one of SEQ ID NOs: 137 and 196; and a CDR-H3 comprising SEQ ID NO: 255;

(iii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 4 and 63; a CDR-H2 comprising one of SEQ ID NOs: 122 and 181; and a CDR-H3 comprising SEQ ID NO: 240;

(iv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 5 and 64; a CDR-H2 comprising one of SEQ ID NOs: 123 and 182; and a CDR-H3 comprising SEQ ID NO: 241;

(v) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 6 and 65; a CDR-H2 comprising one of SEQ ID NOs: 124 and 183; and a CDR-H3 comprising SEQ ID NO: 242;

(vi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 7 and 66; a CDR-H2 comprising one of SEQ ID NOs: 125 and 184; and a CDR-H3 comprising SEQ ID NO: 243;

(vii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 8 and 67; a CDR-H2 comprising one of SEQ ID NOs: 126 and 185; and a CDR-H3 comprising SEQ ID NO: 244;

(viii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 9 and 68; a CDR-H2 comprising one of SEQ ID NOs: 127 and 186; and a CDR-H3 comprising SEQ ID NO: 245;

(ix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 10 and 69; a CDR-H2 comprising one of SEQ ID NOs: 128 and 187; and a CDR-H3 comprising SEQ ID NO: 246;

(x) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 11 and 70; a CDR-H2 comprising one of SEQ ID NOs: 129 and 188; and a CDR-H3 comprising SEQ ID NO: 247;

(xi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 12 and 71; a CDR-H2 comprising one of SEQ ID NOs: 130 and 189; and a CDR-H3 comprising SEQ ID NO: 248;

(xii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 13 and 72; a CDR-H2 comprising one of SEQ ID NOs: 131 and 190; and a CDR-H3 comprising SEQ ID NO: 249;

(xiii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 14 and 73; a CDR-H2 comprising one of SEQ ID NOs: 132 and 191; and a CDR-H3 comprising SEQ ID NO: 250;

(xiv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 15 and 74; a CDR-H2 comprising one of SEQ ID NOs: 133 and 192; and a CDR-H3 comprising SEQ ID NO: 251;

(xv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 16 and 75; a CDR-H2 comprising one of SEQ ID NOs: 134 and 193; and a CDR-H3 comprising SEQ ID NO: 252;

(xvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 17 and 76; a CDR-H2 comprising one of SEQ ID NOs: 135 and 194; and a CDR-H3 comprising SEQ ID NO: 253;

(xvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 18 and 77; a CDR-H2 comprising one of SEQ ID NOs: 136 and 195; and a CDR-H3 comprising SEQ ID NO: 254;

(xviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 20 and 79; a CDR-H2 comprising one of SEQ ID NOs: 138 and 197; and a CDR-H3 comprising SEQ ID NO: 256;

(xix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 21 and 80; a CDR-H2 comprising one of SEQ ID NOs: 139 and 198; and a CDR-H3 comprising SEQ ID NO: 257;

(xx) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 22 and 81; a CDR-H2 comprising one of SEQ ID NOs: 140 and 199; and a CDR-H3 comprising SEQ ID NO: 258;

(xxi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 23 and 82; a CDR-H2 comprising one of SEQ ID NOs: 141 and 200; and a CDR-H3 comprising SEQ ID NO: 259;

(xxii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 24 and 83; a CDR-H2 comprising one of SEQ ID NOs: 142 and 201; and a CDR-H3 comprising SEQ ID NO: 260;

(xxiii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 25 and 84; a CDR-H2 comprising one of SEQ ID NOs: 143 and 202; and a CDR-H3 comprising SEQ ID NO: 261;

(xxiv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 26 and 85; a CDR-H2 comprising one of SEQ ID NOs: 144 and 203; and a CDR-H3 comprising SEQ ID NO: 262;

(xxv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 27 and 86; a CDR-H2 comprising one of SEQ ID NOs: 145 and 204; and a CDR-H3 comprising SEQ ID NO: 263;

(xxvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 28 and 87; a CDR-H2 comprising one of SEQ ID NOs: 146 and 205; and a CDR-H3 comprising SEQ ID NO: 264;

(xxvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 29 and 88; a CDR-H2 comprising one of SEQ ID NOs: 147 and 206; and a CDR-H3 comprising SEQ ID NO: 265;

(xxviii)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 30 and 89; a CDR-H2 comprising one of SEQ ID NOs: 148 and 207; and a CDR-H3 comprising SEQ ID NO: 266;

(xxix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 31 and 90; a CDR-H2 comprising one of SEQ ID NOs: 149 and 208; and a CDR-H3 comprising SEQ ID NO: 267;

(xxx) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 32 and 91; a CDR-H2 comprising one of SEQ ID NOs: 150 and 209; and a CDR-H3 comprising SEQ ID NO: 268;

(xxxi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 33 and 92; a CDR-H2 comprising one of SEQ ID NOs: 151 and 210; and a CDR-H3 comprising SEQ ID NO: 269;

(xxxii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 34 and 93; a CDR-H2 comprising one of SEQ ID NOs: 152 and 211; and a CDR-H3 comprising SEQ ID NO: 270;

(xxxiii)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 35 and 94; a CDR-H2 comprising one of SEQ ID NOs: 153 and 212; and a CDR-H3 comprising SEQ ID NO: 271;

(xxxiv)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 36 and 95; a CDR-H2 comprising one of SEQ ID NOs: 154 and 213; and a CDR-H3 comprising SEQ ID NO: 272;

(xxxv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 37 and 96; a CDR-H2 comprising one of SEQ ID NOs: 155 and 214; and a CDR-H3 comprising SEQ ID NO: 273;

(xxxvi)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 38 and 97; a CDR-H2 comprising one of SEQ ID NOs: 156 and 215; and a CDR-H3 comprising SEQ ID NO: 274;

(xxxvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 39 and 98; a CDR-H2 comprising one of SEQ ID NOs: 157 and 216; and a CDR-H3 comprising SEQ ID NO: 275;

(xxxviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 40 and 99; a CDR-H2 comprising one of SEQ ID NOs: 158 and 217; and a CDR-H3 comprising SEQ ID NO: 276;

(xxxix)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 41 and 100; a CDR-H2 comprising one of SEQ ID NOs: 159 and 218; and a CDR-H3 comprising SEQ ID NO: 277;

(xl) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 42 and 101; a CDR-H2 comprising one of SEQ ID NOs: 160 and 219; and a CDR-H3 comprising SEQ ID NO: 278;

(xli) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 43 and 102; a CDR-H2 comprising one of SEQ ID NOs: 161 and 220; and a CDR-H3 comprising SEQ ID NO: 279;

(xlii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 44 and 103; a CDR-H2 comprising one of SEQ ID NOs: 162 and 221; and a CDR-H3 comprising SEQ ID NO: 280;

(xliii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 45 and 104; a CDR-H2 comprising one of SEQ ID NOs: 163 and 222; and a CDR-H3 comprising SEQ ID NO: 281;

(xliv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 46 and 105; a CDR-H2 comprising one of SEQ ID NOs: 164 and 223; and a CDR-H3 comprising SEQ ID NO: 282;

(xlv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 47 and 106; a CDR-H2 comprising one of SEQ ID NOs: 165 and 224; and a CDR-H3 comprising SEQ ID NO: 283;

(xlvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 48 and 107; a CDR-H2 comprising one of SEQ ID NOs: 166 and 225; and a CDR-H3 comprising SEQ ID NO: 284;

(xlvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 49 and 108; a CDR-H2 comprising one of SEQ ID NOs: 167 and 226; and a CDR-H3 comprising SEQ ID NO: 285;

(xlviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 50 and 109; a CDR-H2 comprising one of SEQ ID NOs: 168 and 227; and a CDR-H3 comprising SEQ ID NO: 286;

(xlix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 51 and 110; a CDR-H2 comprising one of SEQ ID NOs: 169 and 228; and a CDR-H3 comprising SEQ ID NO: 287;

(l) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 52 and 111; a CDR-H2 comprising one of SEQ ID NOs: 170 and 229; and a CDR-H3 comprising SEQ ID NO: 288;

(li) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 53 and 112; a CDR-H2 comprising one of SEQ ID NOs: 171 and 230; and a CDR-H3 comprising SEQ ID NO: 289;

(lii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 54 and 113; a CDR-H2 comprising one of SEQ ID NOs: 172 and 231; and a CDR-H3 comprising SEQ ID NO: 290;

(liii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 55 and 114; a CDR-H2 comprising one of SEQ ID NOs: 173 and 232; and a CDR-H3 comprising SEQ ID NO: 291;

(liv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 56 and 115; a CDR-H2 comprising one of SEQ ID NOs: 174 and 233; and a CDR-H3 comprising SEQ ID NO: 292;

(lv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 57 and 116; a CDR-H2 comprising one of SEQ ID NOs: 175 and 234; and a CDR-H3 comprising SEQ ID NO: 293;

(lvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 59 and 118; a CDR-H2 comprising one of SEQ ID NOs: 177 and 236; and a CDR-H3 comprising SEQ ID NO: 295;

(lvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 60 and 119; a CDR-H2 comprising one of SEQ ID NOs: 178 and 237; and a CDR-H3 comprising SEQ ID NO: 296;

(lviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 61 and 120; a CDR-H2 comprising one of SEQ ID NOs: 179 and 238; and a CDR-H3 comprising SEQ ID NO: 297; or

(lix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 62 and 121; a CDR-H2 comprising one of SEQ ID NOs: 180 and 239; and a CDR-H3 comprising SEQ ID NO: 298.

15. The composition of any one of the preceding claims, wherein the antibody comprises:

(a) a V_L comprising: a CDR-L1 comprising SEQ ID NO: 300; a CDR-L2 comprising SEQ ID NO: 303; and a CDR-L3 comprising SEQ ID NO: 306; or

(b) a V_L comprising: a CDR-L1 comprising SEQ ID NO: 301; a CDR-L2 comprising SEQ ID NO: 304; and a CDR-L3 comprising SEQ ID NO: 307.

16. The composition of any one of the preceding claims, wherein the antibody comprises:

(i) the V_H region is SEQ ID NO: 362, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(ii) the V_H region is SEQ ID NO: 323, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(iii) the V_H region is SEQ ID NO: 308, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(iv) the V_H region is SEQ ID NO: 309, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(v) the V_H region is SEQ ID NO: 310, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(vi) the V_H region is SEQ ID NO: 311, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(vii) the V_H region is SEQ ID NO: 312, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(viii) the V_H region is SEQ ID NO: 313, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(ix) the V_H region is SEQ ID NO: 314, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(x) the V_H region is SEQ ID NO: 315, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xi) the V_H region is SEQ ID NO: 316, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xii) the V_H region is SEQ ID NO: 317, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xiii) the V_H region is SEQ ID NO: 318, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xiv) the V_H region is SEQ ID NO: 319, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xv) the V_H region is SEQ ID NO: 320, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xvi) the V_H region is SEQ ID NO: 321, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xvii) the V_H region is SEQ ID NO: 322, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xviii) the V_H region is SEQ ID NO: 324, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xix) the V_H region is SEQ ID NO: 325, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xx) the V_H region is SEQ ID NO: 326, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof; or

(xxi) the V_H region is SEQ ID NO: 327, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxii) the V_H region is SEQ ID NO: 328, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxiii) the V_H region is SEQ ID NO: 329, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxiv) the V_H region is SEQ ID NO: 330, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxv) the V_H region is SEQ ID NO: 331, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxvi) the V_H region is SEQ ID NO: 332, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxvii) the V_H region is SEQ ID NO: 333, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxviii)the V_H region is SEQ ID NO: 334, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxix) the V_H region is SEQ ID NO: 335, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxx) the V_H region is SEQ ID NO: 336, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxi) the V_H region is SEQ ID NO: 337, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxii) the V_H region is SEQ ID NO: 338, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxiii)the V_H region is SEQ ID NO: 339, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxiv)the V_H region is SEQ ID NO: 340, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxv) the V_H region is SEQ ID NO: 341, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxvi)the V_H region is SEQ ID NO: 342, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxvii) the V_H region is SEQ ID NO: 343, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxviii) the V_H region is SEQ ID NO: 344, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxix)the V_H region is SEQ ID NO: 345, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xl) the V_H region is SEQ ID NO: 346, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xli) the V_H region is SEQ ID NO: 347, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlii) the V_H region is SEQ ID NO: 348, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xliii) the V_H region is SEQ ID NO: 349, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xliv) the V_H region is SEQ ID NO: 350, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlv) the V_H region is SEQ ID NO: 351, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlvi) the V_H region is SEQ ID NO: 352, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlvii) the V_H region is SEQ ID NO: 353, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlviii) the V_H region is SEQ ID NO: 354, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlix) the V_H region is SEQ ID NO: 355, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(l) the V_H region is SEQ ID NO: 356, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(li) the V_H region is SEQ ID NO: 357, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lii) the V_H region is SEQ ID NO: 358, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(liii) the V_H region is SEQ ID NO: 359, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(liv) the V_H region is SEQ ID NO: 360, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lv) the V_H region is SEQ ID NO: 361, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lvi) the V_H region is SEQ ID NO: 363, or a variant thereof, and the V_L region is SEQ ID NO: 368, or a variant thereof;

(lvii) the V_H region is SEQ ID NO: 364, or a variant thereof, and the V_L region is SEQ ID NO: 368, or a variant thereof;

(lviii) the V_H region is SEQ ID NO: 365, or a variant thereof, and the V_L region is SEQ ID NO: 369, or a variant thereof; or

(lix) the V_H region is SEQ ID NO: 366, or a variant thereof, and the V_L region is SEQ ID NO: 369, or a variant thereof.

17. The composition of claim 16, wherein the antibody comprises one or more non-natural amino acids at sites selected from the group of: HC-F404, HC-Y180, and LC-K42 according to the Kabat or EU numbering scheme of Kabat.

18. The composition of claim 16, wherein the antibody comprises a non-natural amino acid at site HC-F404.

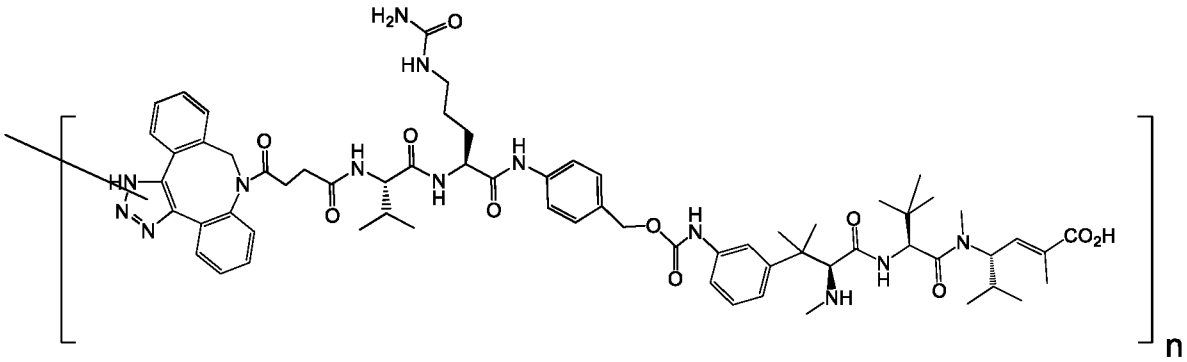
19. The composition of claim 16, wherein the antibody comprises non-natural amino acids at sites HC-F404 and HC-Y180.

20. The composition of claim 16, wherein the antibody comprises non-natural amino acids at sites HC-F404 and LC-K42.

21. The composition of claim 16, wherein the antibody comprises non-natural amino acids at sites HC-Y180 and LC-K42.

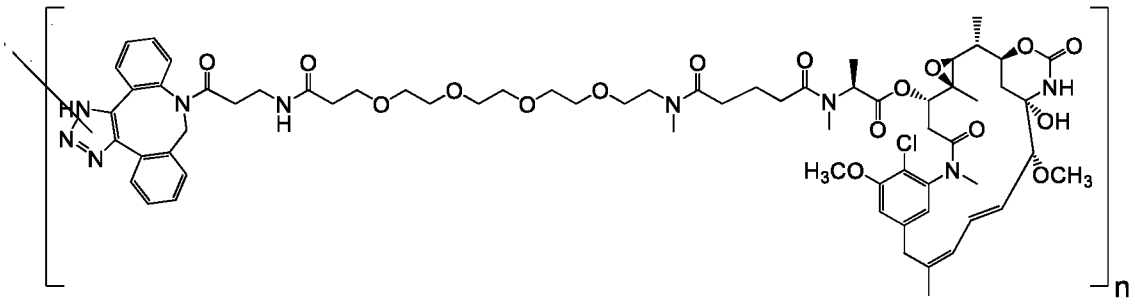
22. The composition of any one of claims 18 to 21, wherein one or both non-natural amino acids is selected from the group consisting of para-azidomethylphenylalanine and p-azido-methyl-L-phenylalanine.

23. The composition of any one of claims 1 to 22, wherein the antibody conjugate has the structure of **Conjugate P**:



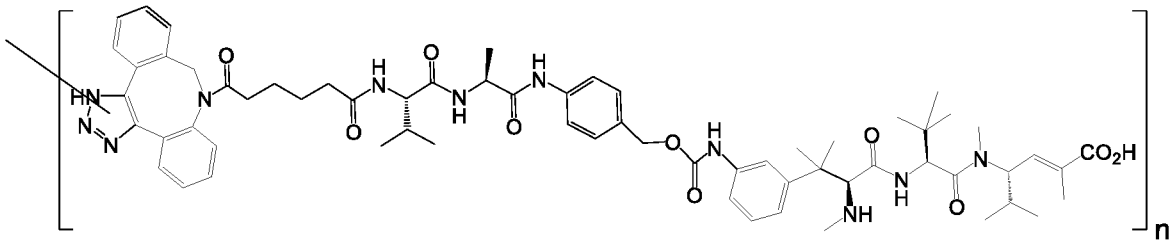
wherein **n** is an integer from 1 to 6.

24. The composition of any one of claims 1 to 22, wherein the antibody conjugate has the structure of **Conjugate M**:



wherein **n** is an integer from 1 to 6.

25. The composition of any one of claims 1 to 22, wherein the antibody conjugate has the structure of **Conjugate Q**:



wherein **n** is an integer from 1 to 6.

26. The composition of any one of claims 1 to 25, wherein the antibody comprises a V_H region of SEQ ID NO: 362, or a variant thereof 7 or fewer amino acid substitutions, and a V_L region of SEQ ID NO: 367, or a variant thereof having 7 or fewer amino acid substitutions.

27. The composition of any one of claims 1 to 25, wherein the antibody comprises a V_H region of SEQ ID NO: 323, or a variant thereof 7 or fewer amino acid substitutions, and a V_L region of SEQ ID NO: 367, or a variant thereof having 7 or fewer amino acid substitutions.

28. The composition of claim 26 or 27, wherein the amino acid substitutions are conservative amino acid substitutions.

29. The composition of any one of the preceding claims, wherein the antibody further comprises at least one constant region domain.

30. The composition of claim 29, wherein the constant region comprises a sequence selected from SEQ ID NOs: 370, 371, and 372.

31. The composition of any one of the preceding claims, wherein the antibody is a monoclonal antibody.

32. The composition of any one of the preceding claims, wherein the antibody is an IgA, an IgD, an IgE, an IgG, or an IgM.

33. The composition of any one of the preceding claims, wherein the antibody is humanized or human.

34. The composition of any one of the preceding claims, wherein the antibody is aglycosylated.

35. The composition of any one of the preceding claims, wherein the antibody is an antibody fragment.

36. The composition of claim 35, wherein the antibody fragment is selected from an Fv fragment, a Fab fragment, a $F(ab')_2$ fragment, a Fab' fragment, an scFv (sFv) fragment, and an scFv-Fc fragment.

37. The composition of claim 36, wherein the antibody fragment is an scFv fragment.

38. The composition of claim 36, wherein the antibody fragment is an scFv-Fc fragment.

39. The composition of any one of the preceding claims, wherein the antibody has a K_D of about 2.26×10^{-11} M to about 7.20×10^{-9} M when bound to human folate receptor at a temperature of 25°C.

40. The composition of any one of the preceding claims, wherein the antibody specifically binds cynomolgus folate receptor.

41. The composition of claim 40, wherein the antibody has a K_D of about 0.19×10^{-9} M to about 2.84×10^{-9} M when bound to cynomolgus folate receptor at a temperature of 25°C.

42. The composition of any one of the preceding claims, wherein the antibody specifically binds mouse folate receptor.

43. The composition of claim 42, wherein the antibody has a K_D of about 0.5×10^{-9} M to about 9.07×10^{-8} M when bound to mouse folate receptor at a temperature of 25°C.

44. A kit comprising composition of any of the preceding claims, and instructions for use of thecomposition.

45. A pharmaceutical composition comprising the composition of any of claims 1 to 43 and a pharmaceutically acceptable carrier.

46. A method of treating or preventing a disease or condition in a subject in need thereof, comprising administering to the subject:

(a) an effective amount of an antibody conjugatecomprising an antibody that specifically binds to folate receptor alpha (FOLR1) linked site-specifically to at least one payload moiety, wherein the antibody comprises one or more non-natural amino acids at sites selected from the group consisting of: HC-F404, HC-K121, HC-Y180, HC-F241, HC-221, LC-T22, LC-S7, LC-N152, LC-K42, LC-E161, LC-D170, HC-S136, HC-S25, HC-A40, HC-S119, HC-S190, HC-K222, HC-R19, HC-Y52, or HC-S70, according to the Kabat, Chothia, or EU numbering scheme; and

(b) one or more PD-1 or PD-L1 inhibitors,.

47. The method of claim 46, wherein the one or more PD-1 or PD-L1 inhibitors comprise a small molecule blocker of the PD-1 or PD-L1 pathway.

48. The method of claim 46, wherein the one or more PD-1 or PD-L1 inhibitors comprise an antibody that inhibits PD-1 or PD-L1 activity.

49. The method of any one of claims 46 to 48, wherein the one or more PD-1 or PD-L1 inhibitors are selected from the group consisting of: CA-170, BMS-8, BMS-202, BMS-936558, CK-301, AUNP12, avelumab, nivolumab, pembrolizumab, atezolizumab, durvalumab, AMP-224, MEDI0680/AMP-514, PDR001, cemiplimab, TSR-042, Tizlelizumab/BGB-A317, CK-301, BMS-936559, camrelizumab, sintilimab, toripalimab, genolimzumab, A167, MGA012, PF-06801591, LY3300054, FAZ053, PD-11, CX-072, BGB-A333, BI 754091, JNJ-63723283, AGEN2034, CA-327, CX-188, STI –A1110, JTX-4014, (LLY) AM0001, CBT-502, FS118, XmAb20717, XmAb23104, AB122, KY1003, RXI-762, PRS-33, ALPN-202, TSR-075, MCLA-145, MGD013, and MGD019.

50. The method of claim 49, wherein the one or more PD-1 or PD-L1 inhibitors comprise avelumab.

51. The method of claim 49, wherein the one or more PD-1 or PD-L1 inhibitors comprise pembrolizumab.

52. The method of claim 48, wherein the antibody that inhibits PD-1 or PD-L1 activity is selected from the antibodies disclosed in WO 2016/077397, WO 2018/156777, and International Application No. PCT/US2013/034213, filed May 23, 2018.

53. The method of any one of claims 46 to 52, wherein the one or more non-natural amino acids is selected from the group consisting of p-acetyl-L-phenylalanine, O-methyl-L-tyrosine, an -3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, a tri-O-acetyl-GlcNAcβ-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-azido-L-phenylalanine, p-azido-methyl-L-phenylalanine, compound 56, p-acyl-L-phenylalanine, p-benzoyl-L-phenylalanine, L-phosphoserine, phosphonoserine, phosphonotyrosine, p-iodo-phenylalanine, p-bromophenylalanine, p-amino-L-phenylalanine, isopropyl-L-phenylalanine, and p-propargyloxy-phenylalanine.

54. The method of claim 53, wherein a residue of the one or more non-natural amino acids is linked to the payload moiety via a linker that is hydrolytically stable.

55. The method of claim 53, wherein a residue of the one or more non-natural amino acids is linked to the payload moiety via a linker that is cleavable.

56. The method of any one of claims 53 to 55, wherein the non-natural amino acid residue is a residue of compound (30) or compound (56).

57. The method of any one of claims 46 to 56, wherein the payload moiety is selected from the group consisting of maytansines, hemiasterlins, amanitins, and auristatins.

58. The method of any one of claims 46 to 57, wherein the payload moiety is selected from the group consisting of DM1, hemiasterlin, amanitin, MMAF, and MMAE.

59. The method of any one of claims 46 to 58, wherein the antibody comprises:

(i) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 58 and 117; a CDR-H2 comprising one of SEQ ID NOs: 176 and 235; and a CDR-H3 comprising SEQ ID NO: 294;

(ii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 19 and 78; a CDR-H2 comprising one of SEQ ID NOs: 137 and 196; and a CDR-H3 comprising SEQ ID NO: 255;

(iii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 4 and 63; a CDR-H2 comprising one of SEQ ID NOs: 122 and 181; and a CDR-H3 comprising SEQ ID NO: 240;

(iv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 5 and 64; a CDR-H2 comprising one of SEQ ID NOs: 123 and 182; and a CDR-H3 comprising SEQ ID NO: 241;

(v) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 6 and 65; a CDR-H2 comprising one of SEQ ID NOs: 124 and 183; and a CDR-H3 comprising SEQ ID NO: 242;

(vi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 7 and 66; a CDR-H2 comprising one of SEQ ID NOs: 125 and 184; and a CDR-H3 comprising SEQ ID NO: 243;

(vii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 8 and 67; a CDR-H2 comprising one of SEQ ID NOs: 126 and 185; and a CDR-H3 comprising SEQ ID NO: 244;

(viii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 9 and 68; a CDR-H2 comprising one of SEQ ID NOs: 127 and 186; and a CDR-H3 comprising SEQ ID NO: 245;

(ix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 10 and 69; a CDR-H2 comprising one of SEQ ID NOs: 128 and 187; and a CDR-H3 comprising SEQ ID NO: 246;

(x) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 11 and 70; a CDR-H2 comprising one of SEQ ID NOs: 129 and 188; and a CDR-H3 comprising SEQ ID NO: 247;

(xi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 12 and 71; a CDR-H2 comprising one of SEQ ID NOs: 130 and 189; and a CDR-H3 comprising SEQ ID NO: 248;

(xii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 13 and 72; a CDR-H2 comprising one of SEQ ID NOs: 131 and 190; and a CDR-H3 comprising SEQ ID NO: 249;

(xiii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 14 and 73; a CDR-H2 comprising one of SEQ ID NOs: 132 and 191; and a CDR-H3 comprising SEQ ID NO: 250;

(xiv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 15 and 74; a CDR-H2 comprising one of SEQ ID NOs: 133 and 192; and a CDR-H3 comprising SEQ ID NO: 251;

(xv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 16 and 75; a CDR-H2 comprising one of SEQ ID NOs: 134 and 193; and a CDR-H3 comprising SEQ ID NO: 252;

(xvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 17 and 76; a CDR-H2 comprising one of SEQ ID NOs: 135 and 194; and a CDR-H3 comprising SEQ ID NO: 253;

(xvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 18 and 77; a CDR-H2 comprising one of SEQ ID NOs: 136 and 195; and a CDR-H3 comprising SEQ ID NO: 254;

(xviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 20 and 79; a CDR-H2 comprising one of SEQ ID NOs: 138 and 197; and a CDR-H3 comprising SEQ ID NO: 256;

(xix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 21 and 80; a CDR-H2 comprising one of SEQ ID NOs: 139 and 198; and a CDR-H3 comprising SEQ ID NO: 257;

(xx) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 22 and 81; a CDR-H2 comprising one of SEQ ID NOs: 140 and 199; and a CDR-H3 comprising SEQ ID NO: 258;

(xxi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 23 and 82; a CDR-H2 comprising one of SEQ ID NOs: 141 and 200; and a CDR-H3 comprising SEQ ID NO: 259;

(xxii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 24 and 83; a CDR-H2 comprising one of SEQ ID NOs: 142 and 201; and a CDR-H3 comprising SEQ ID NO: 260;

(xxiii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 25 and 84; a CDR-H2 comprising one of SEQ ID NOs: 143 and 202; and a CDR-H3 comprising SEQ ID NO: 261;

(xxiv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 26 and 85; a CDR-H2 comprising one of SEQ ID NOs: 144 and 203; and a CDR-H3 comprising SEQ ID NO: 262;

(xxv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 27 and 86; a CDR-H2 comprising one of SEQ ID NOs: 145 and 204; and a CDR-H3 comprising SEQ ID NO: 263;

(xxvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 28 and 87; a CDR-H2 comprising one of SEQ ID NOs: 146 and 205; and a CDR-H3 comprising SEQ ID NO: 264;

(xxvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 29 and 88; a CDR-H2 comprising one of SEQ ID NOs: 147 and 206; and a CDR-H3 comprising SEQ ID NO: 265;

(xxviii)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 30 and 89; a CDR-H2 comprising one of SEQ ID NOs: 148 and 207; and a CDR-H3 comprising SEQ ID NO: 266;

(xxix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 31 and 90; a CDR-H2 comprising one of SEQ ID NOs: 149 and 208; and a CDR-H3 comprising SEQ ID NO: 267;

(xxx) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 32 and 91; a CDR-H2 comprising one of SEQ ID NOs: 150 and 209; and a CDR-H3 comprising SEQ ID NO: 268;

(xxxi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 33 and 92; a CDR-H2 comprising one of SEQ ID NOs: 151 and 210; and a CDR-H3 comprising SEQ ID NO: 269;

(xxxii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 34 and 93; a CDR-H2 comprising one of SEQ ID NOs: 152 and 211; and a CDR-H3 comprising SEQ ID NO: 270;

(xxxiii)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 35 and 94; a CDR-H2 comprising one of SEQ ID NOs: 153 and 212; and a CDR-H3 comprising SEQ ID NO: 271;

(xxxiv)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 36 and 95; a CDR-H2 comprising one of SEQ ID NOs: 154 and 213; and a CDR-H3 comprising SEQ ID NO: 272;

(xxxv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 37 and 96; a CDR-H2 comprising one of SEQ ID NOs: 155 and 214; and a CDR-H3 comprising SEQ ID NO: 273;

(xxxvi)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 38 and 97; a CDR-H2 comprising one of SEQ ID NOs: 156 and 215; and a CDR-H3 comprising SEQ ID NO: 274;

(xxxvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 39 and 98; a CDR-H2 comprising one of SEQ ID NOs: 157 and 216; and a CDR-H3 comprising SEQ ID NO: 275;

(xxxviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 40 and 99; a CDR-H2 comprising one of SEQ ID NOs: 158 and 217; and a CDR-H3 comprising SEQ ID NO: 276;

(xxxix)a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 41 and 100; a CDR-H2 comprising one of SEQ ID NOs: 159 and 218; and a CDR-H3 comprising SEQ ID NO: 277;

(xl) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 42 and 101; a CDR-H2 comprising one of SEQ ID NOs: 160 and 219; and a CDR-H3 comprising SEQ ID NO: 278;

(xli) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 43 and 102; a CDR-H2 comprising one of SEQ ID NOs: 161 and 220; and a CDR-H3 comprising SEQ ID NO: 279;

(xlii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 44 and 103; a CDR-H2 comprising one of SEQ ID NOs: 162 and 221; and a CDR-H3 comprising SEQ ID NO: 280;

(xliii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 45 and 104; a CDR-H2 comprising one of SEQ ID NOs: 163 and 222; and a CDR-H3 comprising SEQ ID NO: 281;

(xliv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 46 and 105; a CDR-H2 comprising one of SEQ ID NOs: 164 and 223; and a CDR-H3 comprising SEQ ID NO: 282;

(xlv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 47 and 106; a CDR-H2 comprising one of SEQ ID NOs: 165 and 224; and a CDR-H3 comprising SEQ ID NO: 283;

(xlvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 48 and 107; a CDR-H2 comprising one of SEQ ID NOs: 166 and 225; and a CDR-H3 comprising SEQ ID NO: 284;

(xlvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 49 and 108; a CDR-H2 comprising one of SEQ ID NOs: 167 and 226; and a CDR-H3 comprising SEQ ID NO: 285;

(xlviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 50 and 109; a CDR-H2 comprising one of SEQ ID NOs: 168 and 227; and a CDR-H3 comprising SEQ ID NO: 286;

(xlix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 51 and 110; a CDR-H2 comprising one of SEQ ID NOs: 169 and 228; and a CDR-H3 comprising SEQ ID NO: 287;

(l) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 52 and 111; a CDR-H2 comprising one of SEQ ID NOs: 170 and 229; and a CDR-H3 comprising SEQ ID NO: 288;

(li) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 53 and 112; a CDR-H2 comprising one of SEQ ID NOs: 171 and 230; and a CDR-H3 comprising SEQ ID NO: 289;

(lii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 54 and 113; a CDR-H2 comprising one of SEQ ID NOs: 172 and 231; and a CDR-H3 comprising SEQ ID NO: 290;

(liii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 55 and 114; a CDR-H2 comprising one of SEQ ID NOs: 173 and 232; and a CDR-H3 comprising SEQ ID NO: 291;

(liv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 56 and 115; a CDR-H2 comprising one of SEQ ID NOs: 174 and 233; and a CDR-H3 comprising SEQ ID NO: 292;

(lv) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 57 and 116; a CDR-H2 comprising one of SEQ ID NOs: 175 and 234; and a CDR-H3 comprising SEQ ID NO: 293;

(lvi) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 59 and 118; a CDR-H2 comprising one of SEQ ID NOs: 177 and 236; and a CDR-H3 comprising SEQ ID NO: 295;

(lvii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 60 and 119; a CDR-H2 comprising one of SEQ ID NOs: 178 and 237; and a CDR-H3 comprising SEQ ID NO: 296;

(lviii) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 61 and 120; a CDR-H2 comprising one of SEQ ID NOs: 179 and 238; and a CDR-H3 comprising SEQ ID NO: 297; or

(lix) a V_H comprising: a CDR-H1 comprising one of SEQ ID NOs: 62 and 121; a CDR-H2 comprising one of SEQ ID NOs: 180 and 239; and a CDR-H3 comprising SEQ ID NO: 298.

60. The method of any one of claims 46 to 59, wherein the antibody comprises:

(a) a V_L comprising: a CDR-L1 comprising SEQ ID NO: 300; a CDR-L2 comprising SEQ ID NO: 303; and a CDR-L3 comprising SEQ ID NO: 306; or

(b) a V_L comprising: a CDR-L1 comprising SEQ ID NO: 301; a CDR-L2 comprising SEQ ID NO: 304; and a CDR-L3 comprising SEQ ID NO: 307.

61. The method of any one of claims 46 to 60, wherein the antibody comprises:

(i) the V_H region is SEQ ID NO: 362, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(ii) the V_H region is SEQ ID NO: 323, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(iii) the V_H region is SEQ ID NO: 308, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(iv) the V_H region is SEQ ID NO: 309, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(v) the V_H region is SEQ ID NO: 310, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(vi) the V_H region is SEQ ID NO: 311, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(vii) the V_H region is SEQ ID NO: 312, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(viii) the V_H region is SEQ ID NO: 313, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(ix) the V_H region is SEQ ID NO: 314, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(x) the V_H region is SEQ ID NO: 315, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xi) the V_H region is SEQ ID NO: 316, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xii) the V_H region is SEQ ID NO: 317, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xiii) the V_H region is SEQ ID NO: 318, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xiv) the V_H region is SEQ ID NO: 319, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xv) the V_H region is SEQ ID NO: 320, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xvi) the V_H region is SEQ ID NO: 321, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xvii) the V_H region is SEQ ID NO: 322, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xviii) the V_H region is SEQ ID NO: 324, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xix) the V_H region is SEQ ID NO: 325, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xx) the V_H region is SEQ ID NO: 326, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof; or

(xxi) the V_H region is SEQ ID NO: 327, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxii) the V_H region is SEQ ID NO: 328, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxiii) the V_H region is SEQ ID NO: 329, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxiv) the V_H region is SEQ ID NO: 330, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxv) the V_H region is SEQ ID NO: 331, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxvi) the V_H region is SEQ ID NO: 332, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxvii) the V_H region is SEQ ID NO: 333, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxviii)the V_H region is SEQ ID NO: 334, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxix) the V_H region is SEQ ID NO: 335, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxx) the V_H region is SEQ ID NO: 336, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxi) the V_H region is SEQ ID NO: 337, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxii) the V_H region is SEQ ID NO: 338, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxiii)the V_H region is SEQ ID NO: 339, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxiv)the V_H region is SEQ ID NO: 340, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxv) the V_H region is SEQ ID NO: 341, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxvi)the V_H region is SEQ ID NO: 342, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxvii) the V_H region is SEQ ID NO: 343, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxviii) the V_H region is SEQ ID NO: 344, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xxxix)the V_H region is SEQ ID NO: 345, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xl) the V_H region is SEQ ID NO: 346, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xli) the V_H region is SEQ ID NO: 347, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlii) the V_H region is SEQ ID NO: 348, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xliii) the V_H region is SEQ ID NO: 349, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xliv) the V_H region is SEQ ID NO: 350, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlv) the V_H region is SEQ ID NO: 351, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlvi) the V_H region is SEQ ID NO: 352, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlvii) the V_H region is SEQ ID NO: 353, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlviii) the V_H region is SEQ ID NO: 354, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(xlix) the V_H region is SEQ ID NO: 355, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(l) the V_H region is SEQ ID NO: 356, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(li) the V_H region is SEQ ID NO: 357, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lii) the V_H region is SEQ ID NO: 358, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(liii) the V_H region is SEQ ID NO: 359, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(liv) the V_H region is SEQ ID NO: 360, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lv) the V_H region is SEQ ID NO: 361, or a variant thereof, and the V_L region is SEQ ID NO: 367, or a variant thereof;

(lvi) the V_H region is SEQ ID NO: 363, or a variant thereof, and the V_L region is SEQ ID NO: 368, or a variant thereof;

(lvii) the V_H region is SEQ ID NO: 364, or a variant thereof, and the V_L region is SEQ ID NO: 368, or a variant thereof;

(lviii) the V_H region is SEQ ID NO: 365, or a variant thereof, and the V_L region is SEQ ID NO: 369, or a variant thereof; or

(lix) the V_H region is SEQ ID NO: 366, or a variant thereof, and the V_L region is SEQ ID NO: 369, or a variant thereof.

62. The method of claim 61, wherein the antibody comprises one or more non-natural amino acids at sites selected from the group of: HC-F404, HC-Y180, and LC-K42 according to the Kabat or EU numbering scheme of Kabat.

63. The method of claim 61, wherein the antibody comprises a non-natural amino acid at site HC-F404.

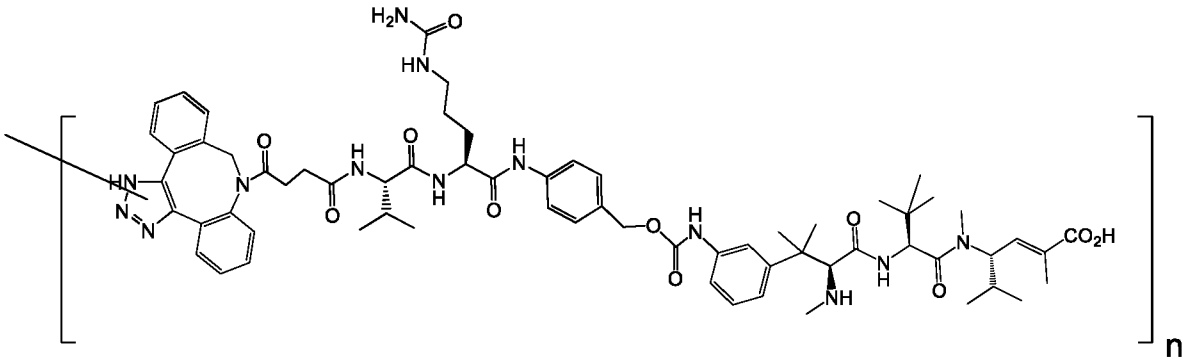
64. The method of claim 61, wherein the antibody comprises non-natural amino acids at sites HC-F404 and HC-Y180.

65. The method of claim 61, wherein the antibody comprises non-natural amino acids at sites HC-F404 and LC-K42.

66. The method of claim 61, wherein the antibody comprises non-natural amino acids at sites HC-Y180 and LC-K42.

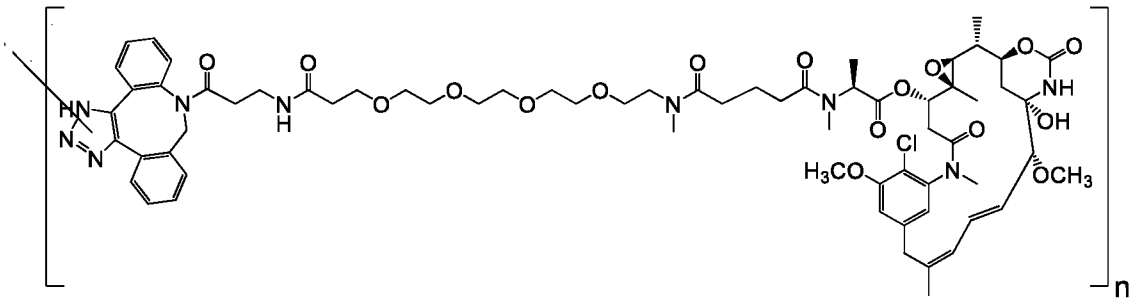
67. The method of any one of claims 63 to 66, wherein one or both non-natural amino acids is selected from the group consisting of para-azidomethylphenylalanine and p-azido-methyl-L-phenylalanine.

68. The method of any one of claims 46 to 67, wherein the antibody conjugate has the structure of **Conjugate P**:



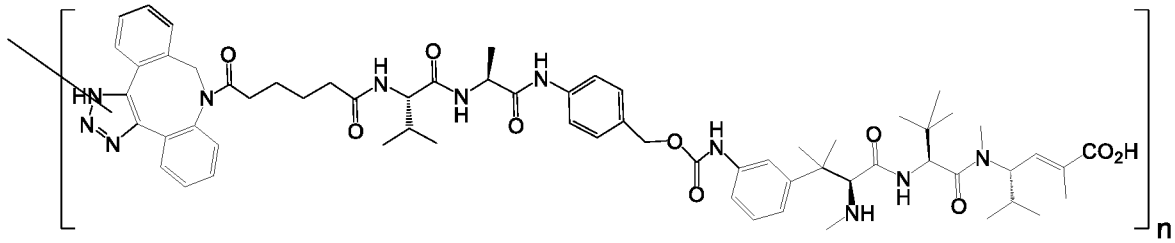
wherein **n** is an integer from 1 to 6.

69. The method of any one of claims 46 to 67, wherein the antibody conjugate has the structure of **Conjugate M**:



wherein **n** is an integer from 1 to 6.

70. The method of any one of claims 46 to 67, wherein the antibody conjugate has the structure of **Conjugate Q**:



wherein **n** is an integer from 1 to 6.

71. The method of any one of claims 46 to 70, wherein the antibody comprises a V_H region of SEQ ID NO: 362, or a variant thereof 7 or fewer amino acid substitutions, and a V_L region of SEQ ID NO: 367, or a variant thereof having 7 or fewer amino acid substitutions.

72. The method of any one of claims 46 to 70, wherein the antibody comprises a V_H region of SEQ ID NO: 323, or a variant thereof 7 or fewer amino acid substitutions, and a V_L region of SEQ ID NO: 367, or a variant thereof having 7 or fewer amino acid substitutions.

73. The method of claim 71 or 72, wherein the amino acid substitutions are conservative amino acid substitutions.

74. The method of any one of the claims 46 to 73, wherein the antibody further comprises at least one constant region domain.

75. The method of claim 74, wherein the constant region comprises a sequence selected from SEQ ID NOs: 370, 371, and 372.

76. The method of any one of claims 46 to 75, wherein the antibody is a monoclonal antibody.

77. The method of any one of claims 46 to 76, wherein the antibody is an IgA, an IgD, an IgE, an IgG, or an IgM.

78. The method of any one of claims 46 to 77, wherein the antibody is humanized or human.

79. The method of any one of claims 46 to 78, wherein the antibody is aglycosylated.

80. The method of any one of claims 46 to 79, wherein the antibody is an antibody fragment.

81. The method of claim 80, wherein the antibody fragment is selected from an Fv fragment, a Fab fragment, a $F(ab')_2$ fragment, a Fab' fragment, an scFv (sFv) fragment, and an scFv-Fc fragment.

82. The method of claim 81, wherein the antibody fragment is an scFv fragment.

83. The method of claim 81, wherein the antibody fragment is an scFv-Fc fragment.

84. The method of any one of claims 46to 83, wherein the disease or condition is a cancer.

85. The method of any one of claims 46 to 84, wherein the disease or condition is breast cancer.

86. The method of any one of claims 46 to 84, wherein the disease or condition is triple-negative breast cancer (TNBC).

87. The method of any one of claims 46 to 84, wherein the disease or condition is ovarian cancer.

88. The method of any one of claims 46 to 84, wherein the disease or condition is lung cancer.

89. The method of any one of claims 46 to 84, wherein the disease or condition is non-small cell lung cancer (NSCLC).

90. The method of any one of claims 46 to 84, wherein the disease or condition is endometrial cancer.

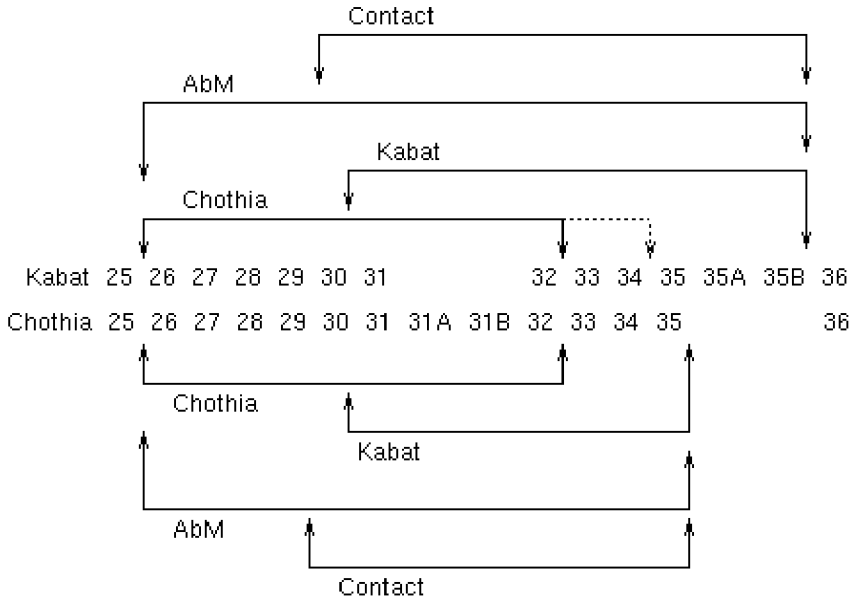

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FIG.1

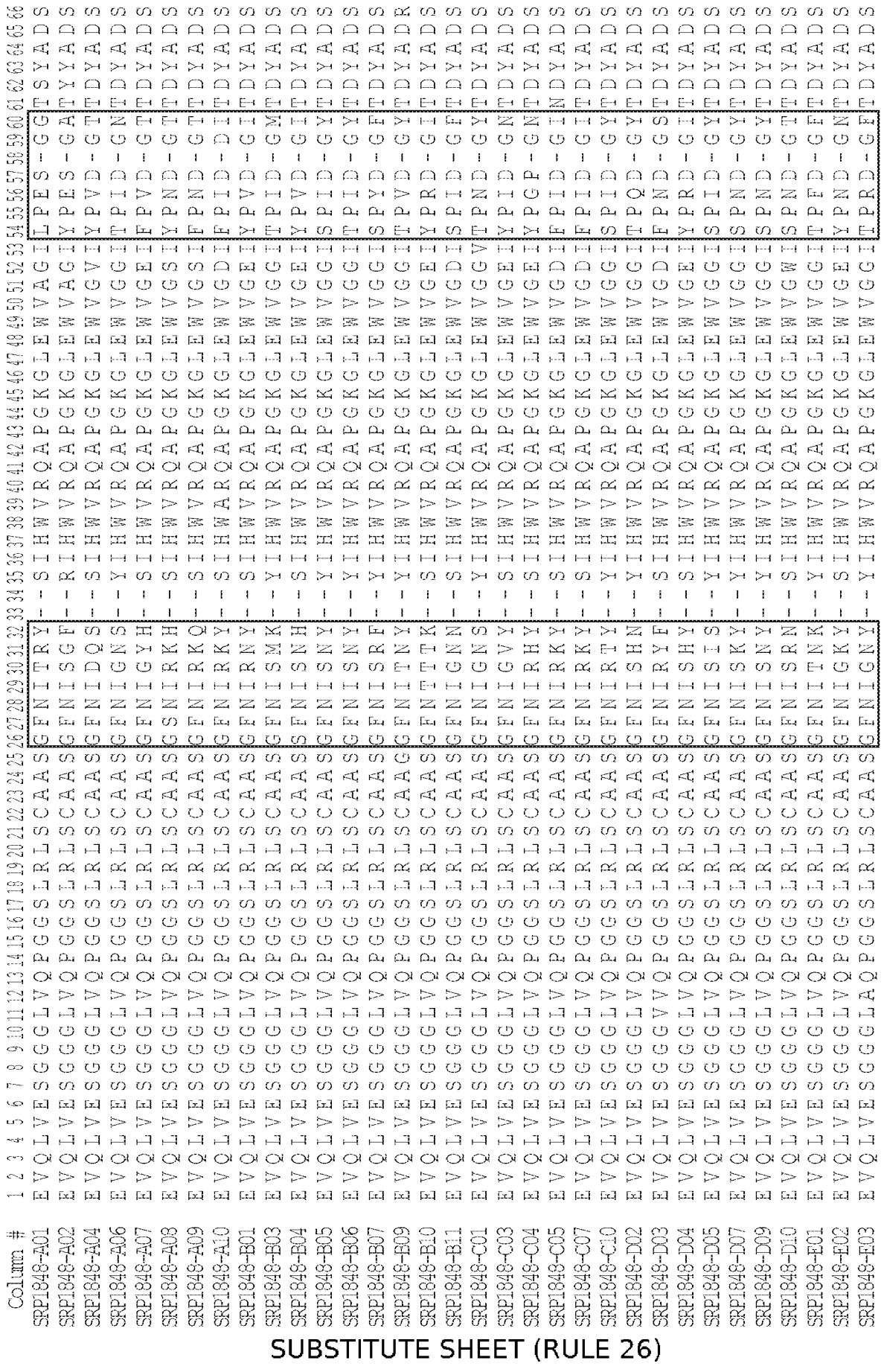

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FIG. 2

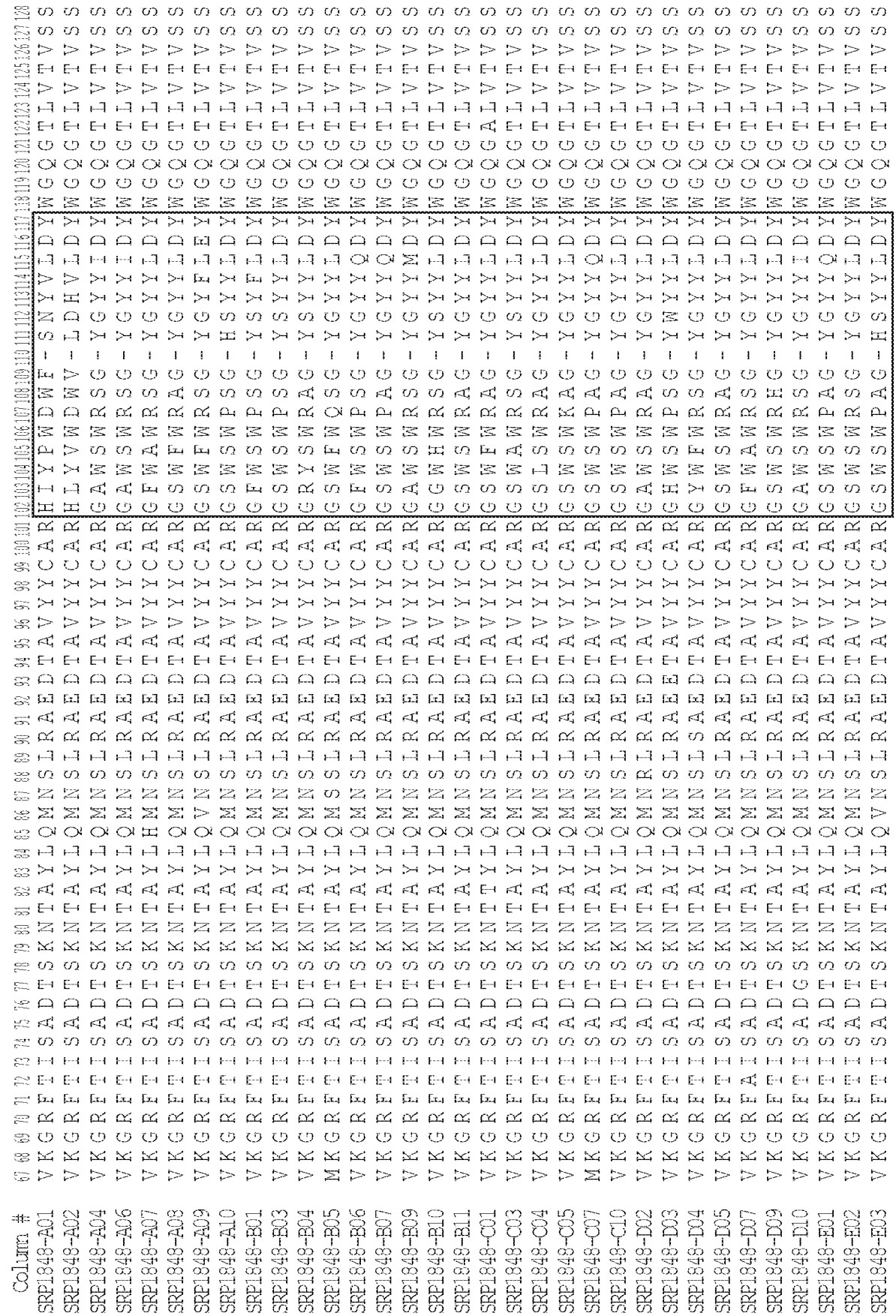

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FIG. 2 (Cont.)

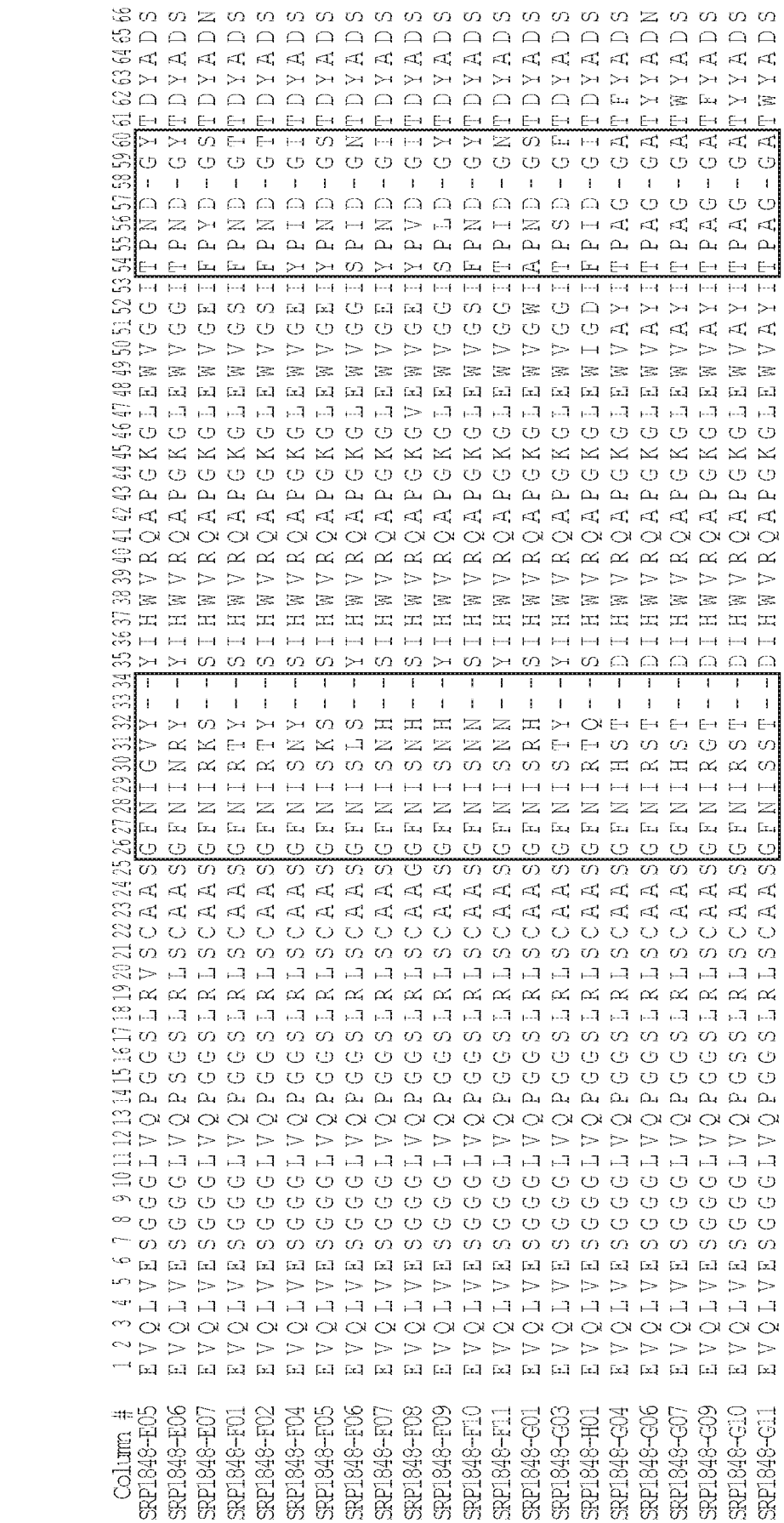

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FIG. 3

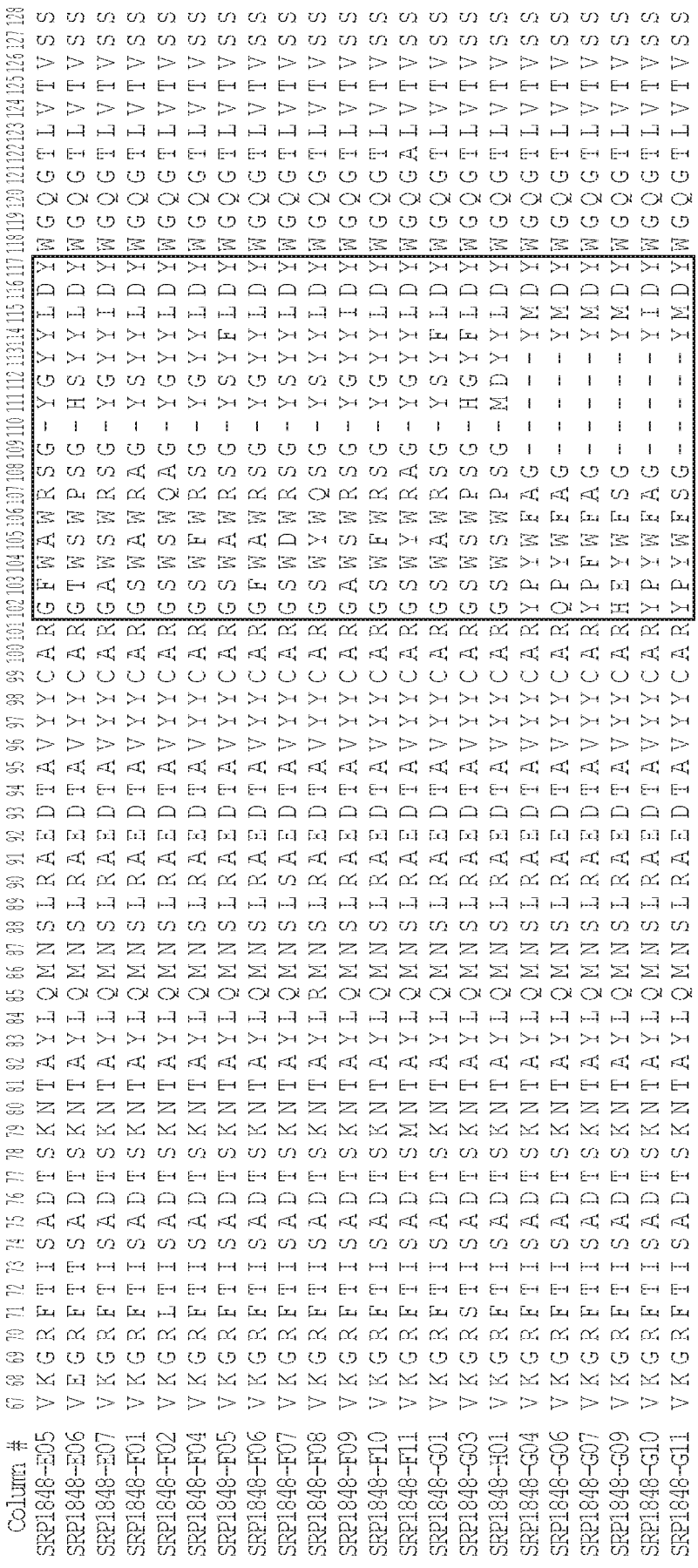

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SRP1848-G04 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R Y P Y W F A G - - - - Y M D Y W G Q G T L V T V S S
SRP1848-G06 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R Q P Y W F A G - - - - Y M D Y W G Q G T L V T V S S
SRP1848-G07 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R Y P F W F A G - - - - Y M D Y W G Q G T L V T V S S
SRP1848-G09 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R H E Y W F S G - - - - Y M D Y W G Q G T L V T V S S
SRP1848-G10 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R Y P Y W F A G - - - - Y I D Y W G Q G T L V T V S S
SRP1848-G11 V K G R F T I S A D T S K N T A Y L Q M N S L R A E D T A V Y Y C A R Y P Y W F S G - - - - Y M D Y W G Q G T L V T V S S

FIG. 3 (Cont.)

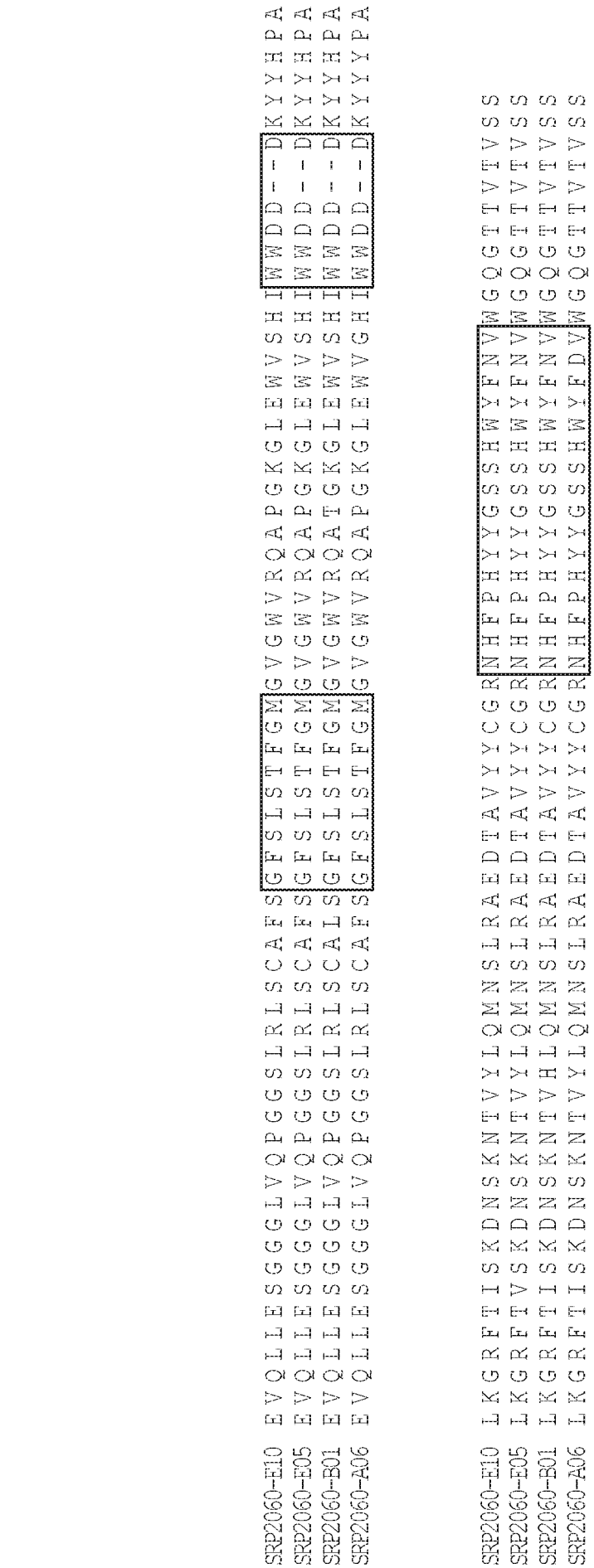

SRP2060-E10 EVQLLESGGGLVQPGGSLRLSCAFSGFSLSTFGMGVGWVRQAPGKGLEWVSHIWWDD--DKYYHPA
SRP2060-E05 EVQLLESGGGLVQPGGSLRLSCAFSGFSLSTFGMGVGWVRQAPGKGLEWVSHIWWDD--DKYYHPA
SRP2060-B01 EVQLLESGGGLVQPGGSLRLSCALSGFSLSTFGMGVGWVRQATGKGLEWVSHIWWDD--DKYYHPA
SRP2060-A06 EVQLLESGGGLVQPGGSLRLSCAFSGFSLSTFGMGVGWVRQAPGKGLEWVSHIWWDD--DKYYPA
SRP2060-E10 LKGRFTISKDNSKNTVYLQMNSLRAEDTAVYYCGRNHFPHYYGSSHWYFNVWGQGTTVTVSS
SRP2060-E05 LKGRFTVSKDNSKNTVYLQMNSIRAEDTAVYYCGRNNHFPHYYGSSHWYFNVWGQGTTVTVSS
SRP2060-B01 LKGRFTISKDNSKNTVHLQMNSLRAEDTAVYYCGRNHFPHYYGSSHWYFNVWGQGTTVTVSS
SRP2060-A06 LKGRFTISKDNSKNTVYLQMNSLRAEDTAVYYCGRNHFPHYYGSSHWYFDVWGQGTTVTVSS

FIG. 4

Column #	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59
TRASTUZUMAB	D	I	Q	M	T	Q	S	P	S	S	L	S	A	S	V	G	D	R	V	T	I	T	C	R	A	S	Q	D	V	N	T	A	V	A	W	Y	Q	Q	K	P	G	K	A	P	K	L	L	I	Y	S	A	S	F	L	Y	S	G	V	P
H6D1-LC4	E	I	V	M	T	Q	S	P	A	T	L	S	L	S	P	G	E	R	A	T	L	S	C	K	A	S	Q	D	I	N	S	Y	L	S	W	Y	Q	Q	K	P	G	Q	A	P	R	L	L	I	Y	R	A	N	R	L	V	D	G	I	P
H6D1-LC5	D	I	Q	M	T	Q	S	P	S	T	L	S	A	S	V	G	D	R	V	T	I	T	C	K	A	S	Q	D	I	N	S	Y	L	S	W	Y	Q	Q	K	P	G	K	A	P	K	L	L	I	Y	R	A	N	R	L	V	D	G	V	P

Column #	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100	101	102	103	104	105	106	107	108
TRASTUZUMAB	S	R	F	S	G	S	R	S	G	T	D	F	T	L	T	I	S	S	L	Q	P	E	D	F	A	T	Y	Y	C	Q	Q	H	Y	T	T	-	P	P	T	F	G	Q	G	T	K	V	E	I	K
H6D1-LC4	A	R	F	S	G	S	G	S	G	T	D	Y	T	L	T	I	S	S	L	E	P	E	D	F	A	V	Y	Y	C	L	Q	-	Y	D	E	F	P	Y	T	F	G	G	G	T	K	V	E	I	K
H6D1-LC5	S	R	F	S	G	S	G	S	G	T	E	F	T	L	T	I	S	S	L	Q	P	D	D	F	A	T	Y	Y	C	L	Q	-	Y	D	E	F	P	Y	T	F	G	G	G	T	K	V	E	I	K

FIG. 5

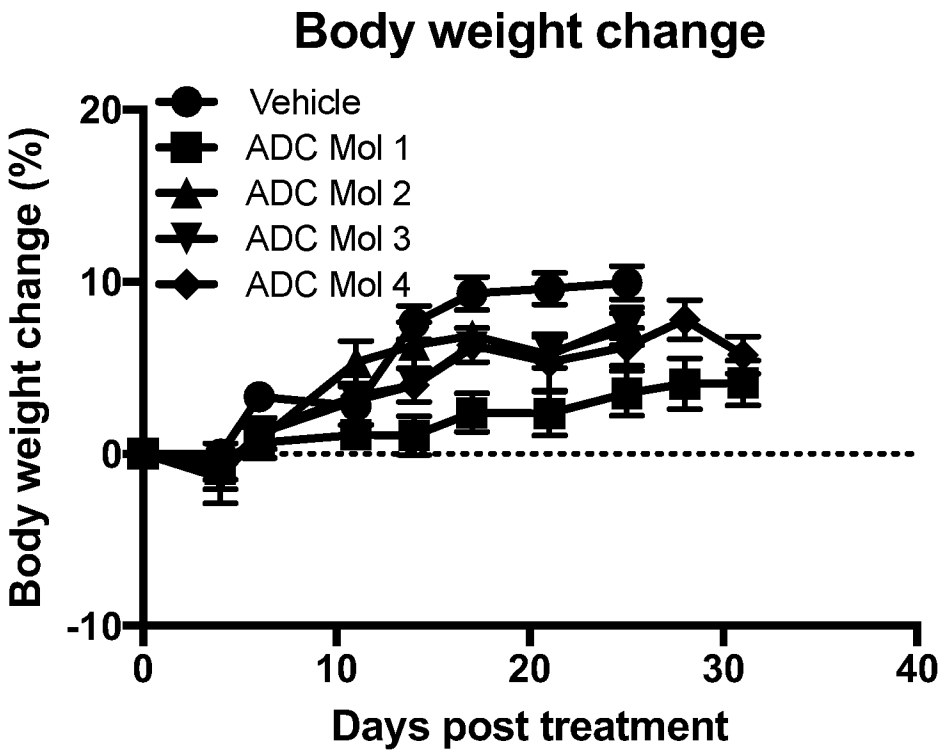

Body weight change
Vehicle
ADC Mol 1
ADC Mol 2
ADC Mol 3
ADC Mol 4
Body weight change (%)
20
10
0
-10
0
10
20
30
40
Days post treatment

FIG. 6

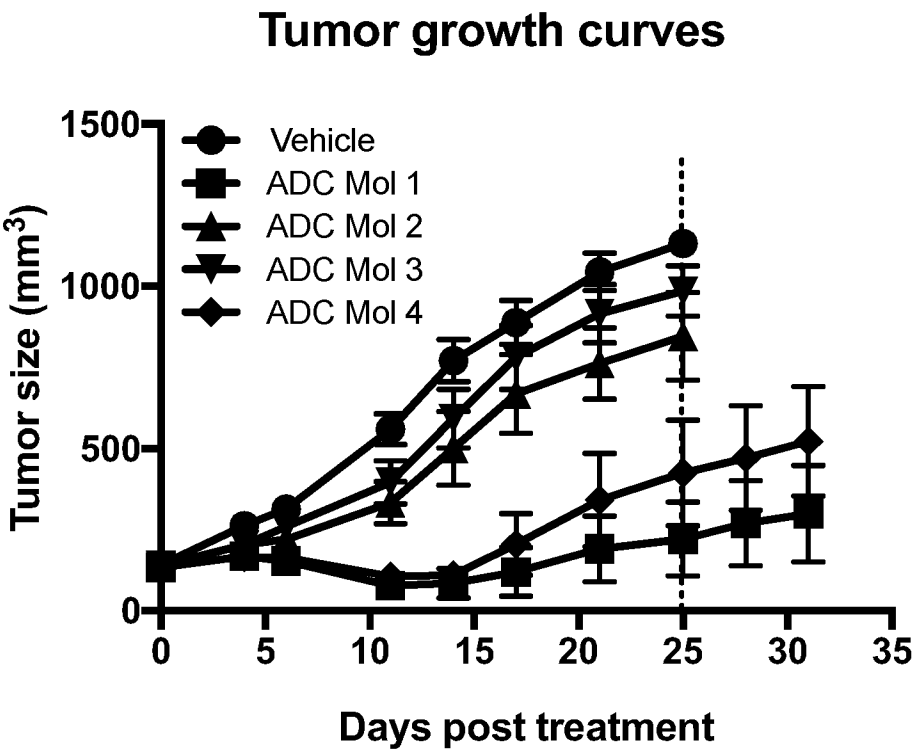

Tumor growth curves
Vehicle
ADC Mol 1
ADC Mol 2
ADC Mol 3
ADC Mol 4
Tumor size (mm3)
1500
1000
500
0
0
5
10
15
20
25
30
35
Days post treatment

FIG. 7A

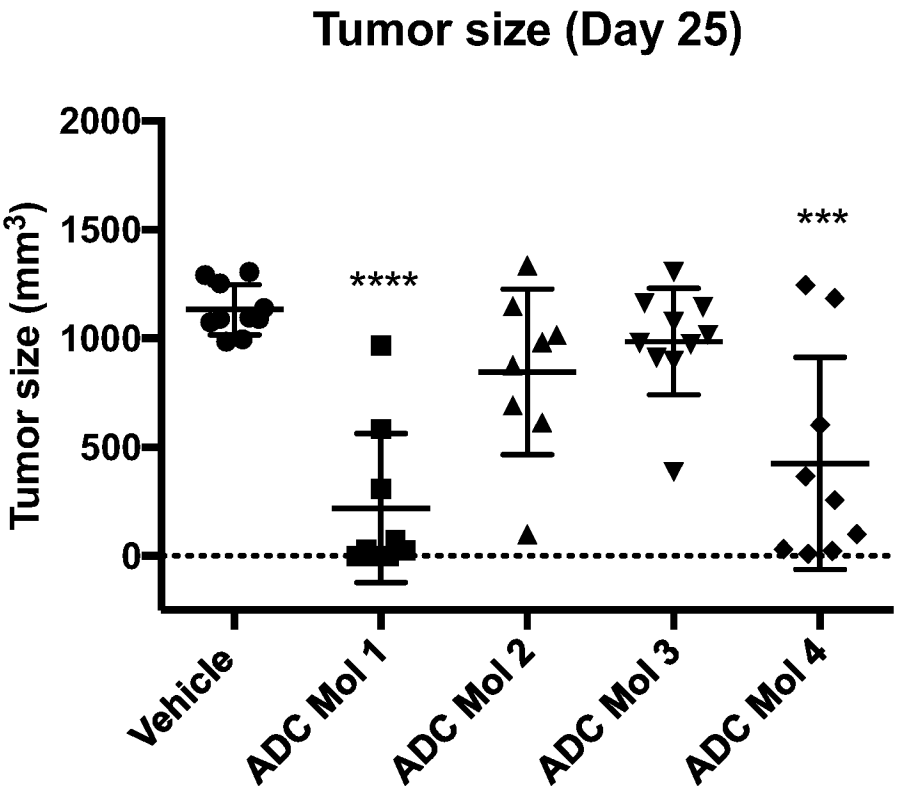

Tumor size (Day 25)
Tumor size (mm3)
2000
1500
1000
500
0
****
***
Vehicle
ADC Mol 1
ADC Mol 2
ADC Mol 3
ADC Mol 4

FIG. 7B

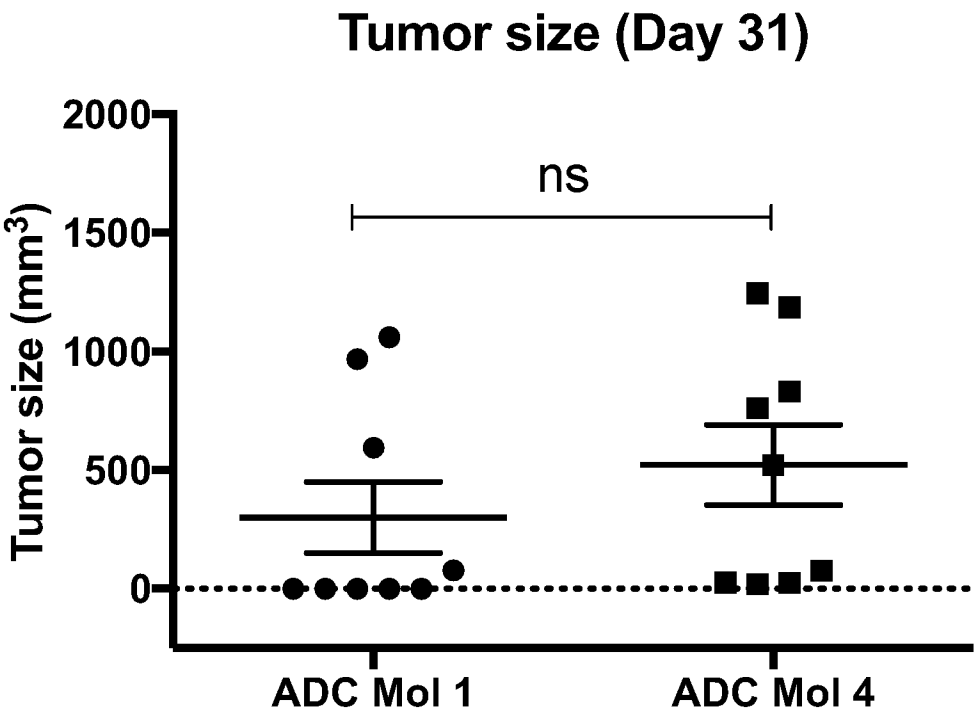

Tumor size (Day 31)
2000
1500
1000
500
0
Tumor size (mm3)
ns
ADC Mol 1
ADC Mol 4

FIG. 8

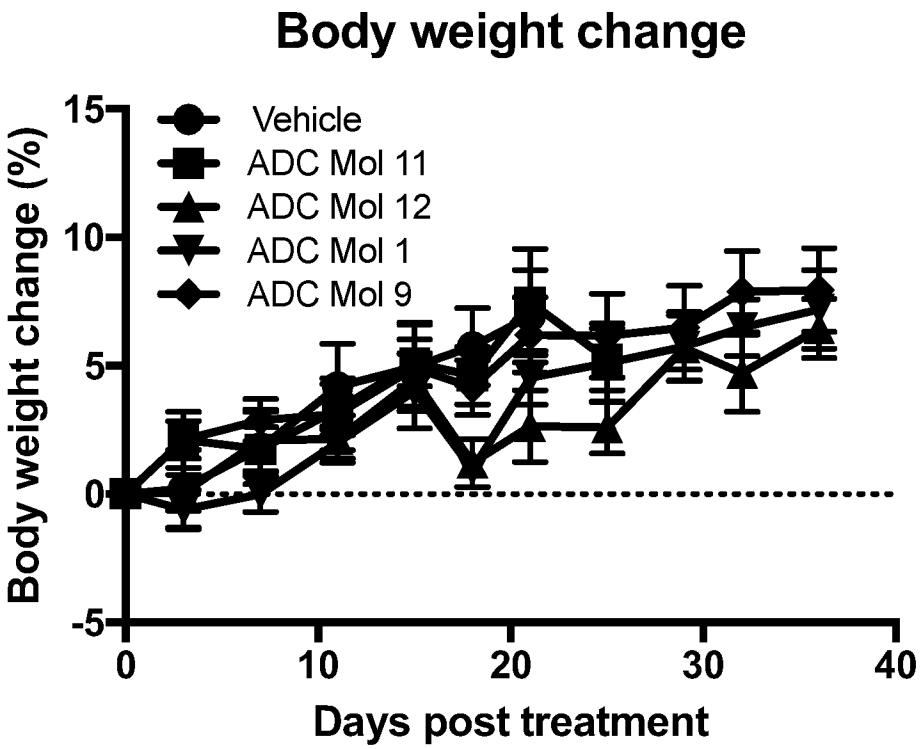

Body weight change
Vehicle
ADC Mol 11
ADC Mol 12
ADC Mol 1
ADC Mol 9
Body weight change (%)
15
10
5
0
-5
0
10
20
30
40
Days post treatment

FIG. 9

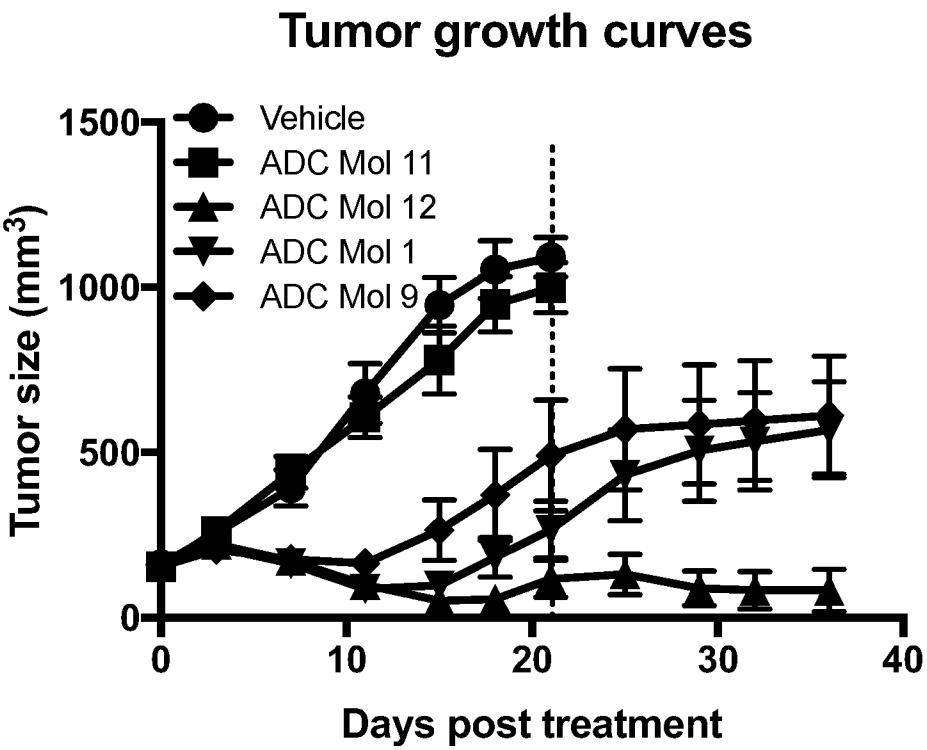

Tumor growth curves
Vehicle
ADC Mol 11
ADC Mol 12
ADC Mol 1
ADC Mol 9
Tumor size (mm3)
1500
1000
500
0
0
10
20
30
40
Days post treatment

FIG. 10A

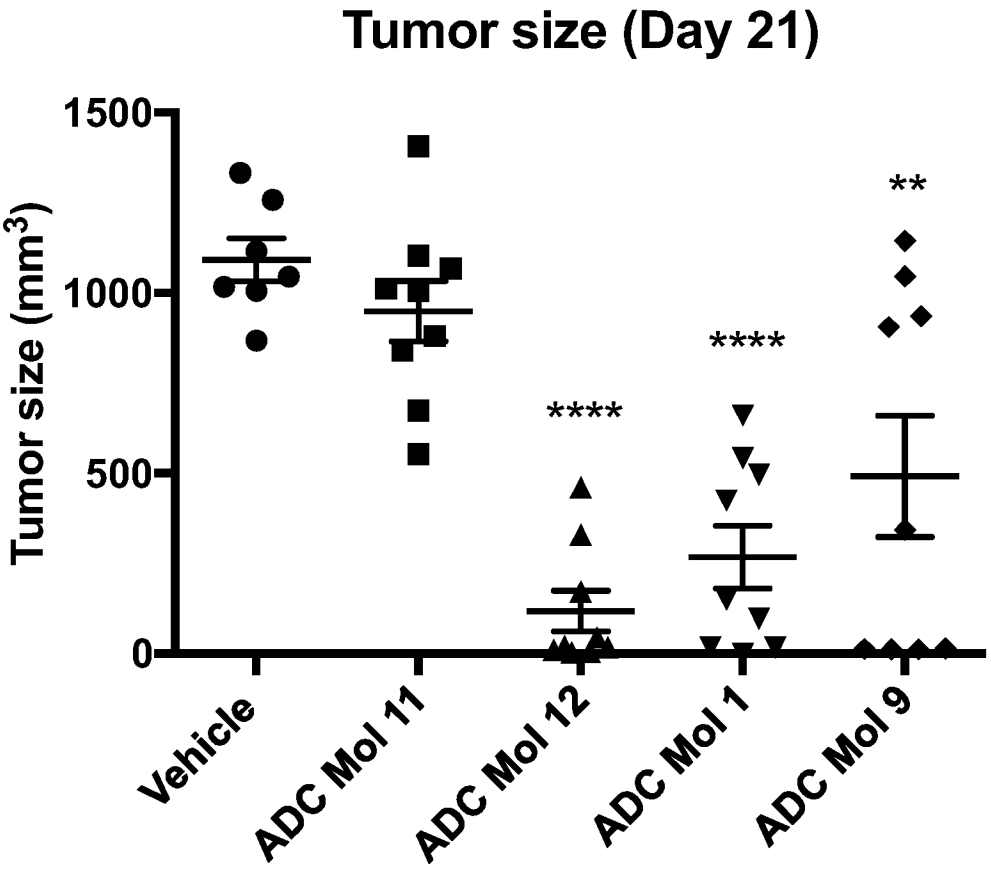

Tumor size (Day 21)
Tumor size (mm3)
1500
1000
500
0
****
****
**
Vehicle
ADC Mol 11
ADC Mol 12
ADC Mol 1
ADC Mol 9

FIG. 10B

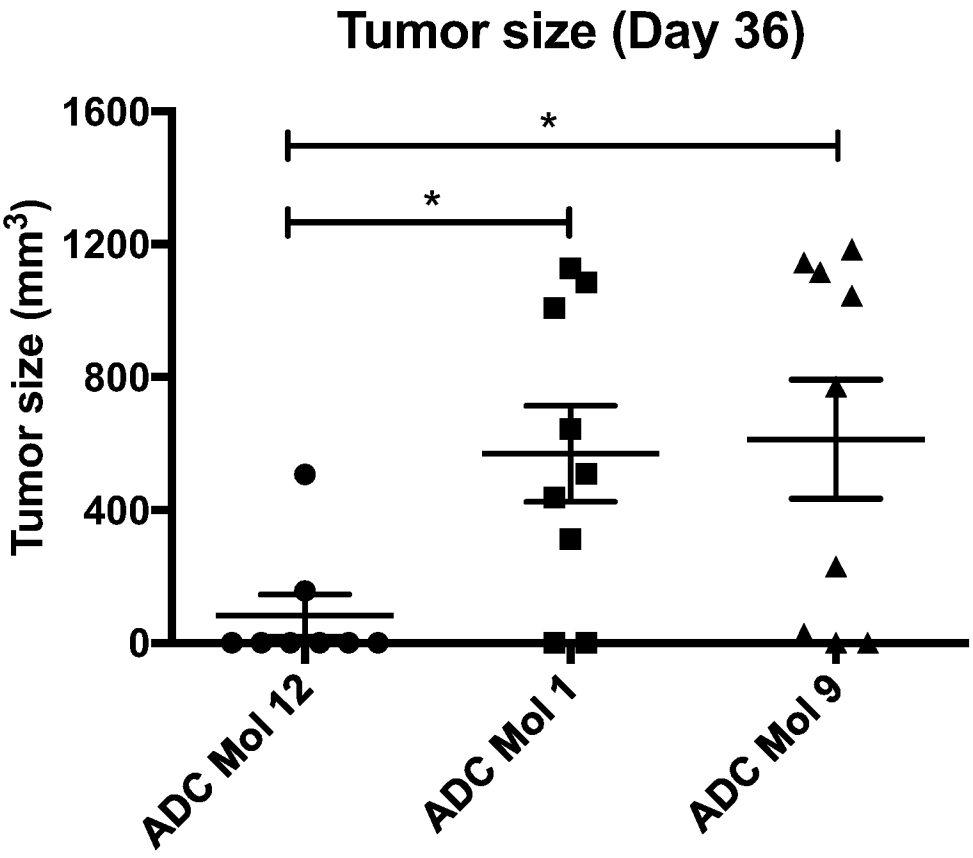

Tumor size (Day 36)
Tumor size (mm3)
1600
1200
800
400
0
*
*
ADC Mol 12
ADC Mol 1
ADC Mol 9

FIG. 11

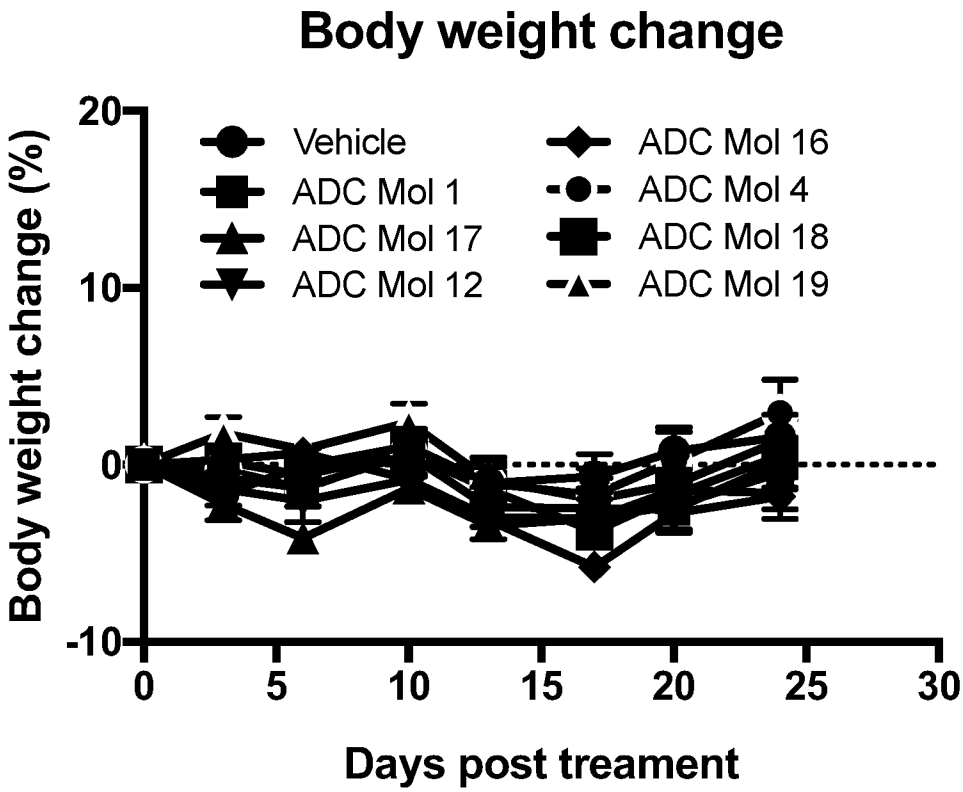

Body weight change
Body weight change (%)
20
10
0
-10
0
5
10
15
20
25
30
Days post treament
Vehicle
ADC Mol 1
ADC Mol 17
ADC Mol 12
ADC Mol 16
ADC Mol 4
ADC Mol 18
ADC Mol 19

FIG. 12

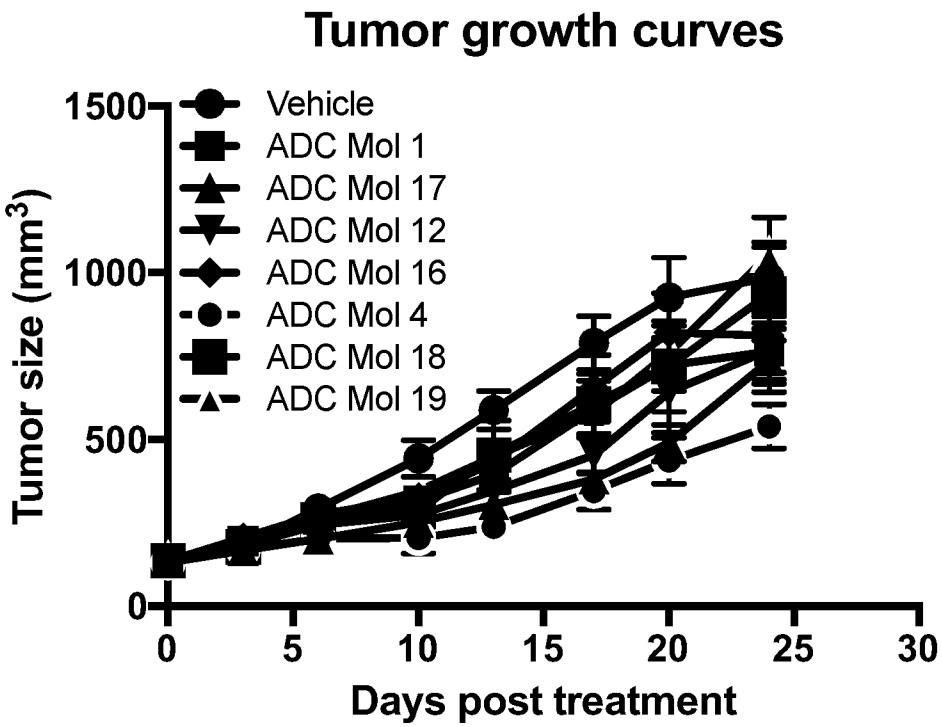

Tumor growth curves
Vehicle
ADC Mol 1
ADC Mol 17
ADC Mol 12
ADC Mol 16
ADC Mol 4
ADC Mol 18
ADC Mol 19
Tumor size (mm3)
1500
1000
500
0
0
5
10
15
20
25
30
Days post treatment

FIG. 13A

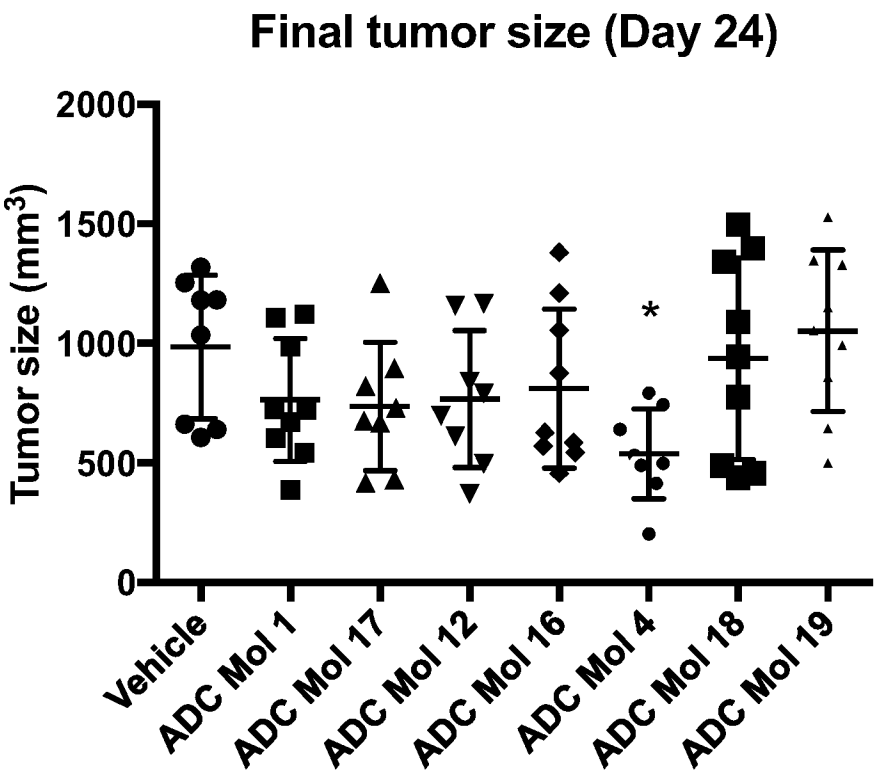

Final tumor size (Day 24)
Tumor size (mm3)
2000
1500
1000
500
0
*
Vehicle
ADC Mol 1
ADC Mol 17
ADC Mol 12
ADC Mol 16
ADC Mol 4
ADC Mol 18
ADC Mol 19

FIG. 13B

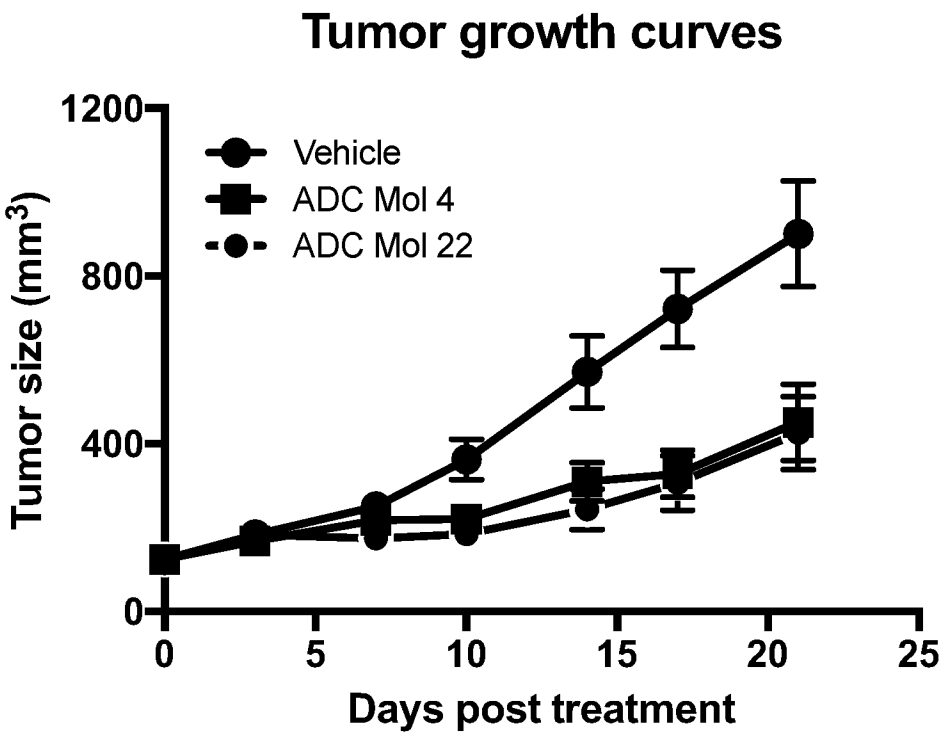

Tumor growth curves
1200
800
400
0
Tumor size (mm3)
Vehicle
ADC Mol 4
ADC Mol 22
0
5
10
15
20
25
Days post treatment

FIG. 14A

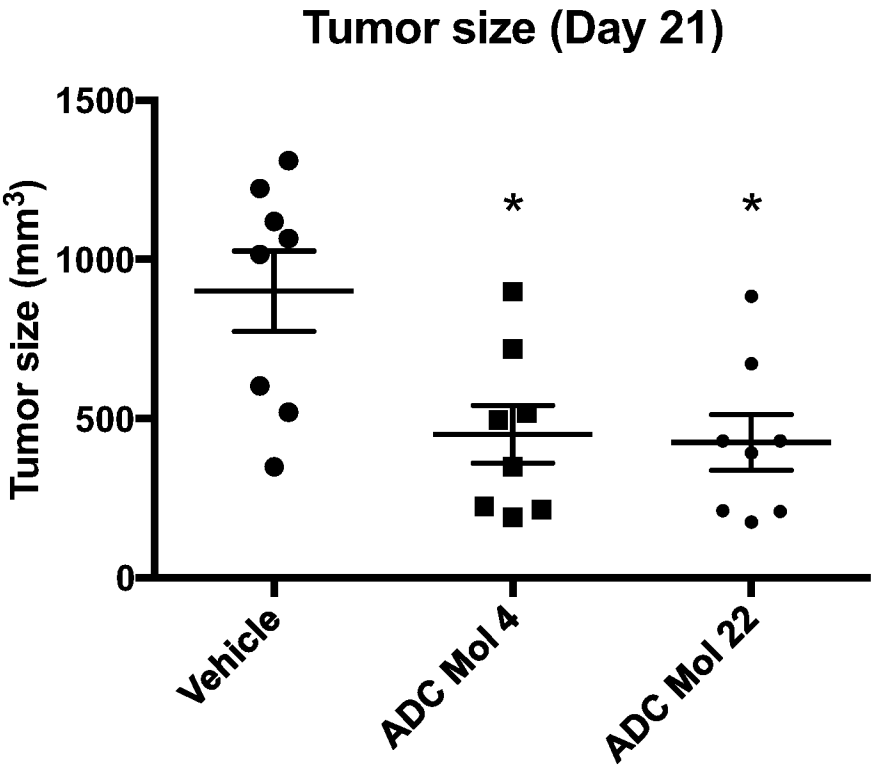

Tumor size (Day 21)
Tumor size (mm3)
1500
1000
500
0
*
*
Vehicle
ADC Mol 4
ADC Mol 22

FIG. 14B

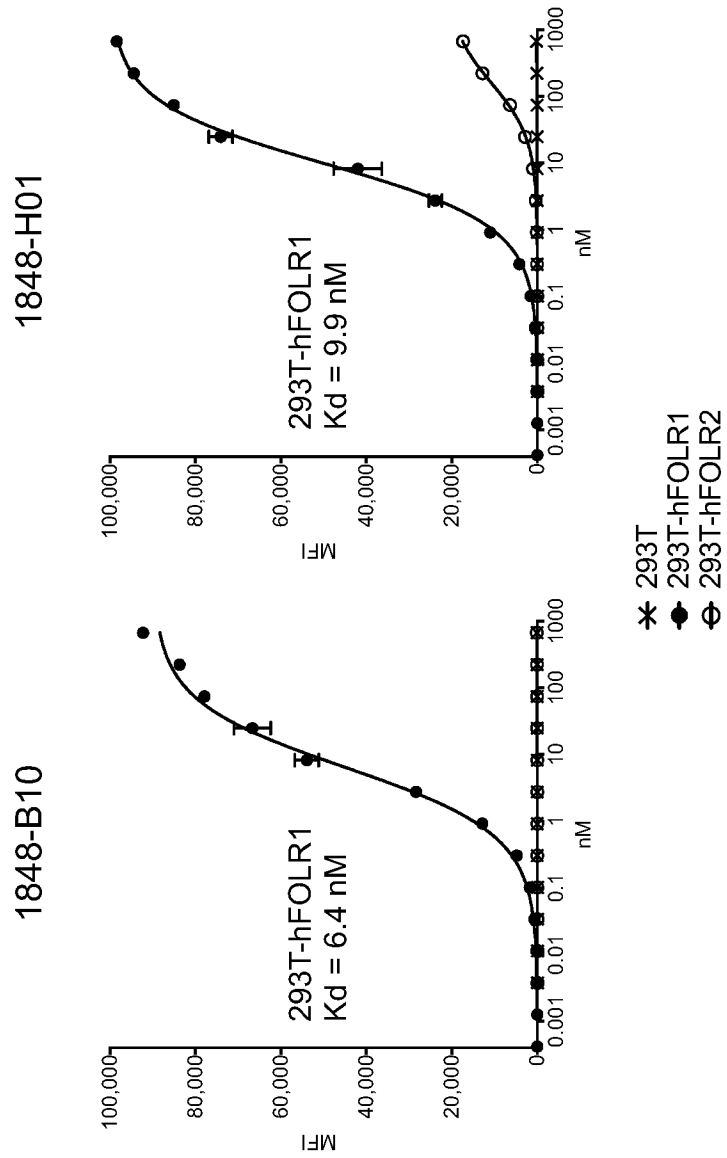

1848-H01
293T-hFOLR1
Kd = 9.9 nM
1848-B10
293T-hFOLR1
Kd = 6.4 nM
MFI
nM
293T
293T-hFOLR1
293T-hFOLR2

FIG. 15

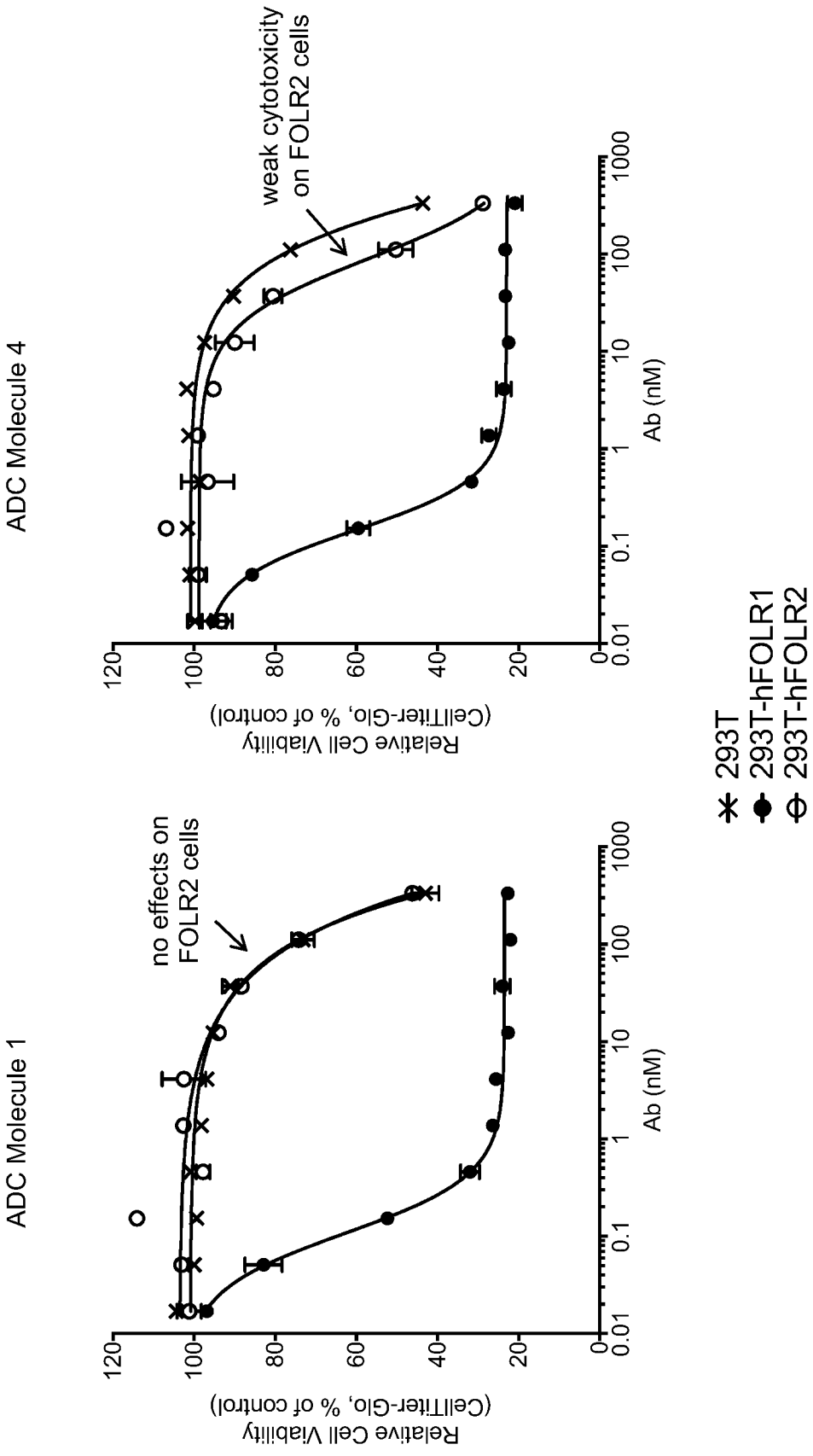

ADC Molecule 4
ADC Molecule 1
weak cytotoxicity on FOLR2 cells
no effects on FOLR2 cells
Relative Cell Viability (CellTiter-Glo, % of control)
Ab (nM)
293T
293T-hFOLR1
293T-hFOLR2

FIG. 16

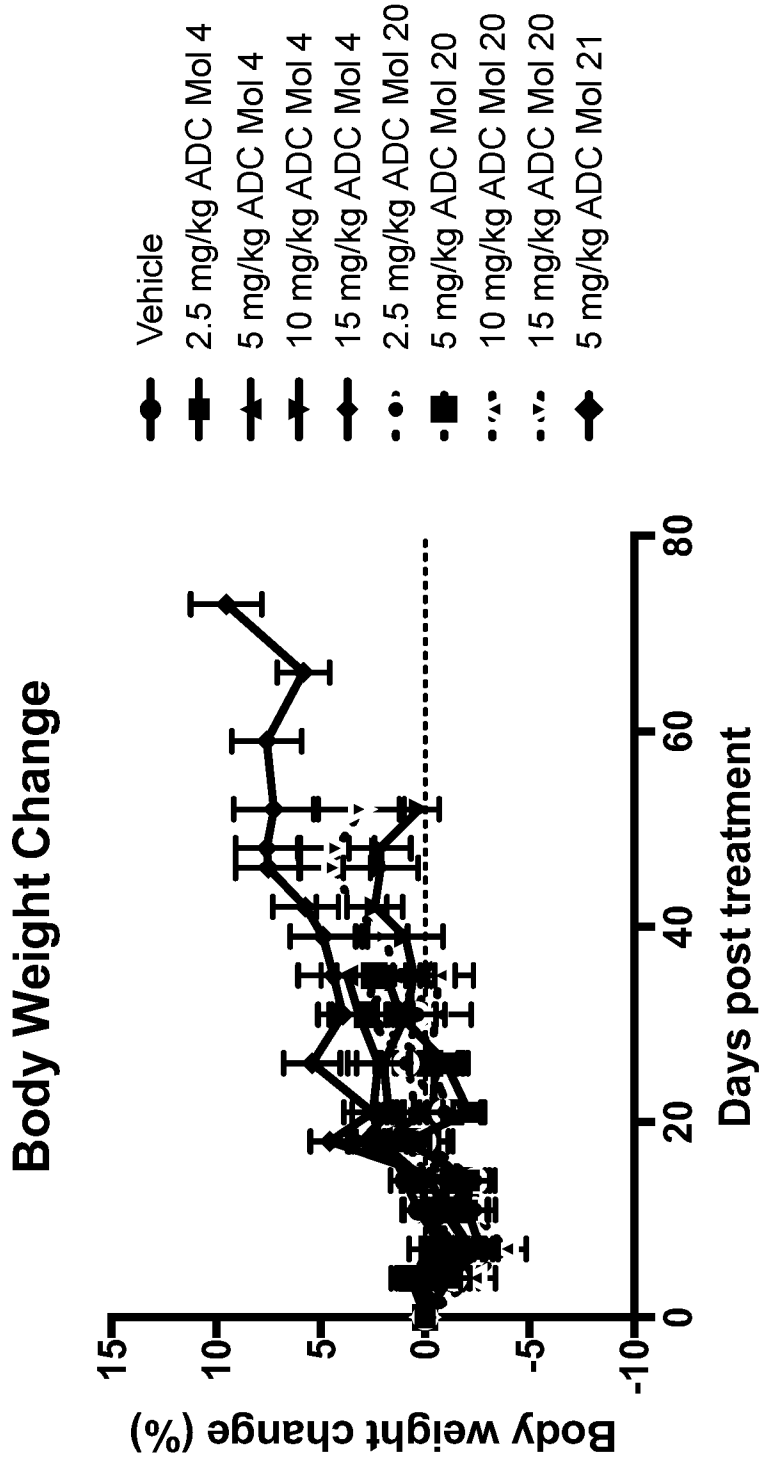

Body Weight Change
Body weight change (%)
15
10
5
0
-5
-10
0
20
40
60
80
Days post treatment
Vehicle
2.5 mg/kg ADC Mol 4
5 mg/kg ADC Mol 4
10 mg/kg ADC Mol 4
15 mg/kg ADC Mol 4
2.5 mg/kg ADC Mol 20
5 mg/kg ADC Mol 20
10 mg/kg ADC Mol 20
15 mg/kg ADC Mol 20
5 mg/kg ADC Mol 21

FIG. 17

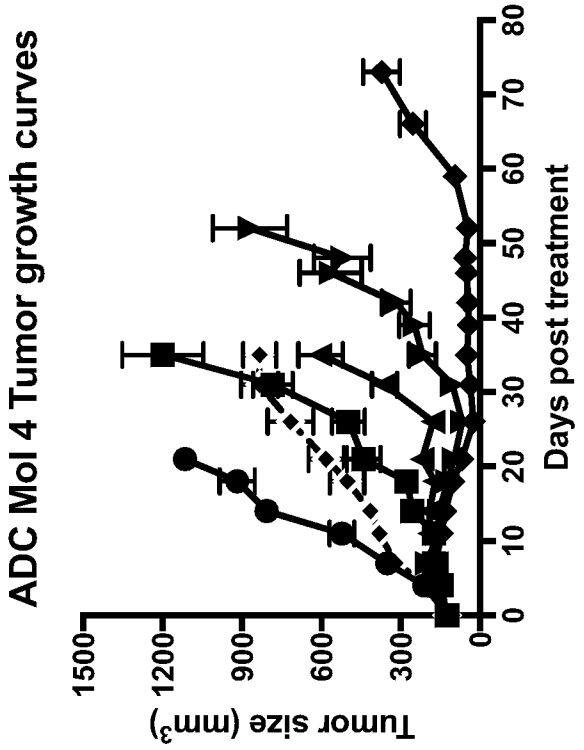

ADC Mol 4 Tumor growth curves
Tumor size (mm3)
1500
1200
900
600
300
0
0
10
20
30
40
50
60
70
80
Days post treatment
Vehicle
2.5 mg/kg ADC Mol 4
5 mg/kg ADC Mol 4
10 mg/kg ADC Mol 4
15 mg/kg ADC Mol 4
5 mg/kg ADC Mol 21

FIG. 18A

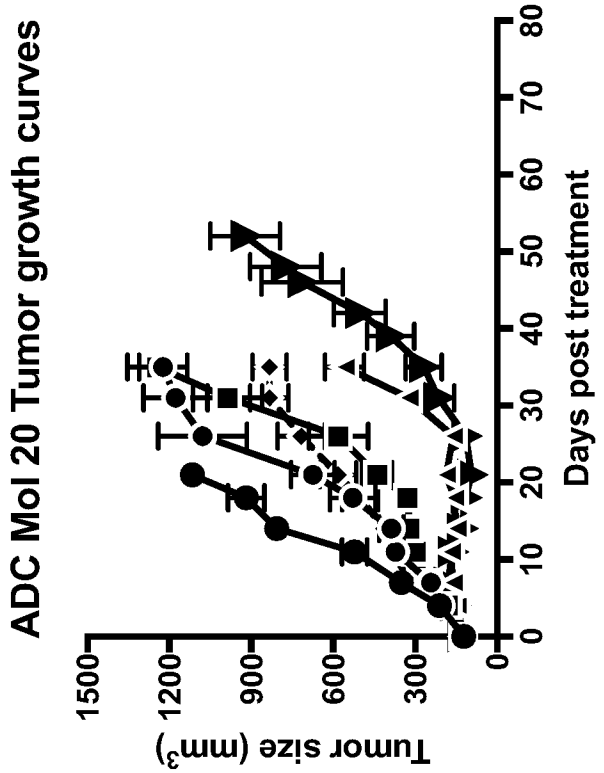

ADC Mol 20 Tumor growth curves
Tumor size (mm3)
1500
1200
900
600
300
0
0
10
20
30
40
50
60
70
80
Days post treatment
Vehicle
2.5 mg/kg ADC Mol 20
5 mg/kg ADC Mol 20
10 mg/kg ADC Mol 20
15 mg/kg ADC Mol 20
5 mg/kg ADC Mol 21

FIG. 18B

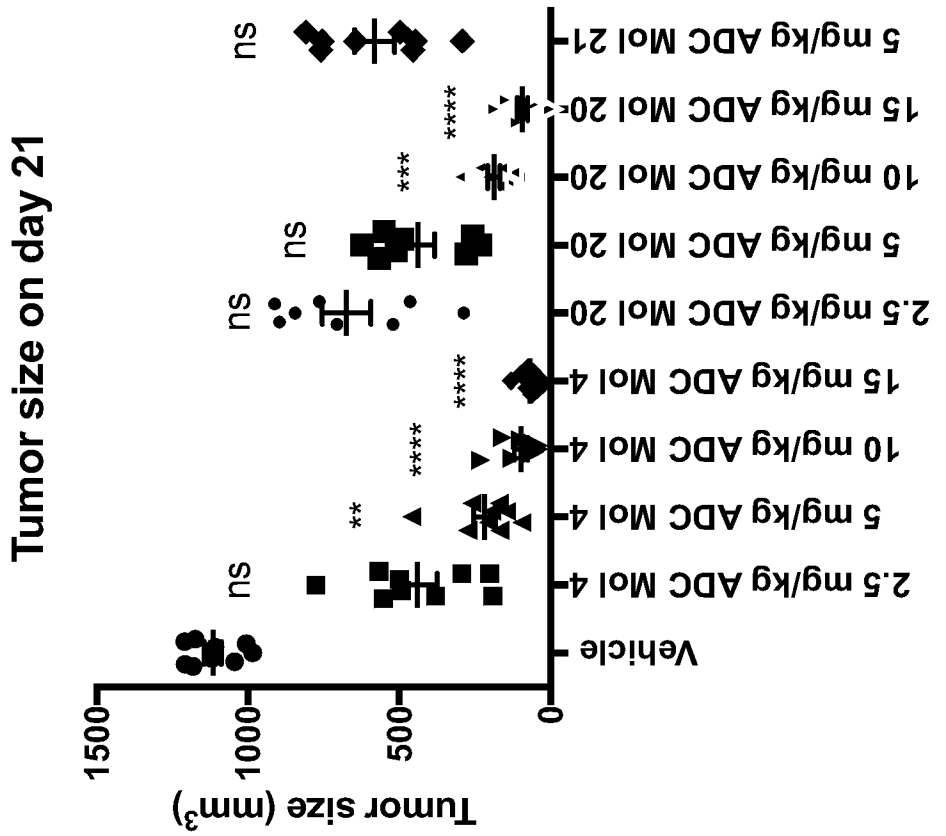

Tumor size on day 21
Tumor size (mm3)
1500
1000
500
0
Vehicle
2.5 mg/kg ADC Mol 4
5 mg/kg ADC Mol 4
10 mg/kg ADC Mol 4
15 mg/kg ADC Mol 4
2.5 mg/kg ADC Mol 20
5 mg/kg ADC Mol 20
10 mg/kg ADC Mol 20
15 mg/kg ADC Mol 20
5 mg/kg ADC Mol 21
ns
**
****
****
ns
ns
***
****
ns

FIG. 18C

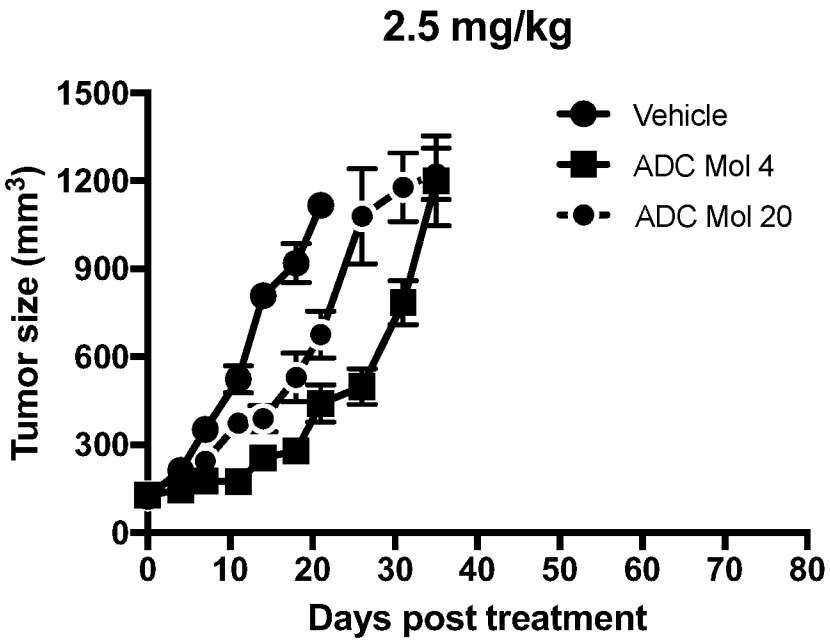


FIG. 19A

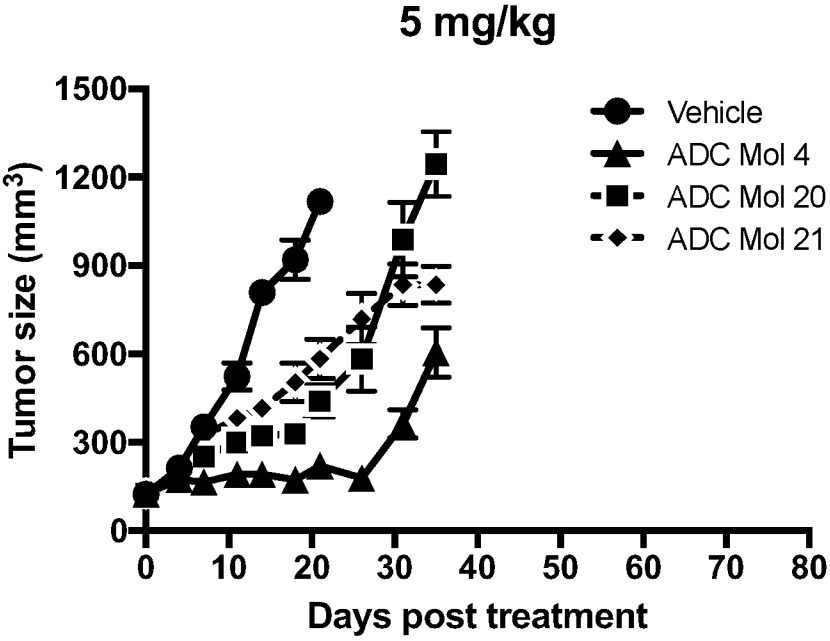


FIG. 19B

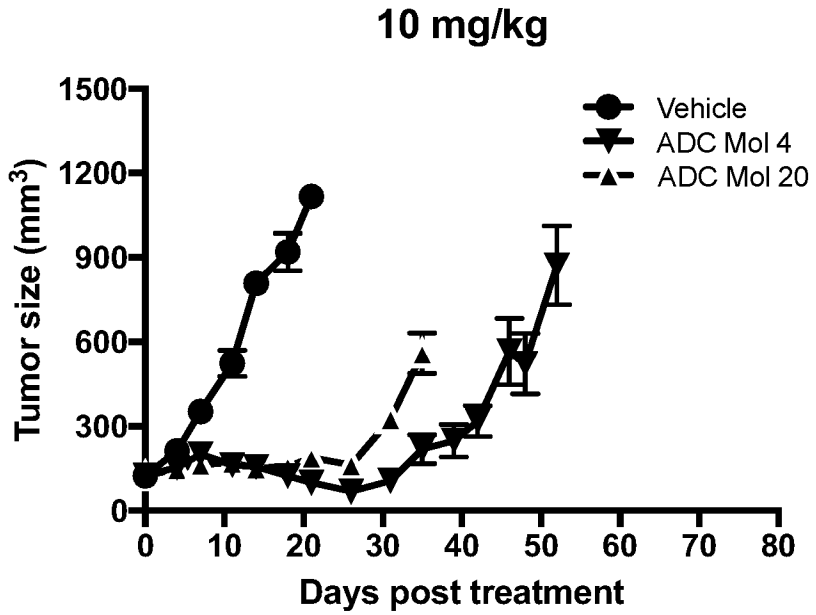


FIG. 19C

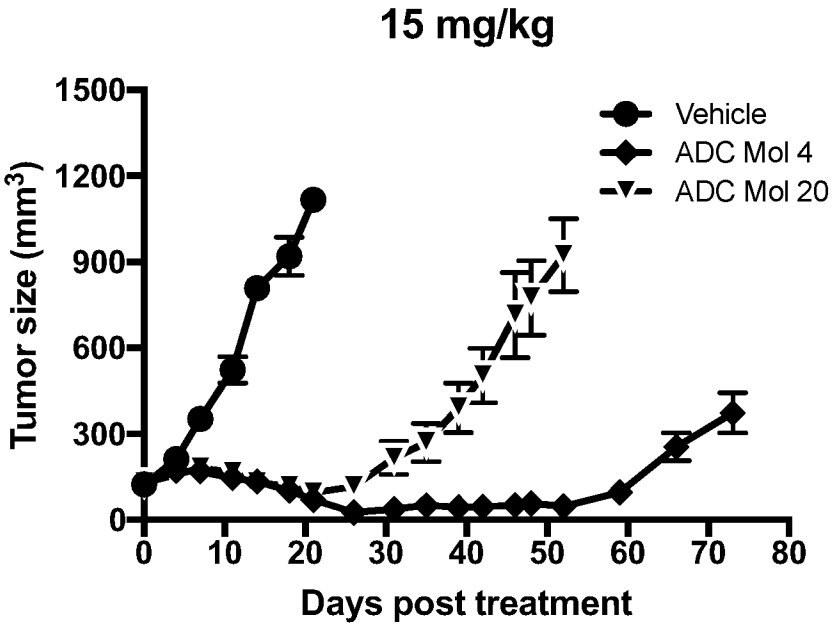
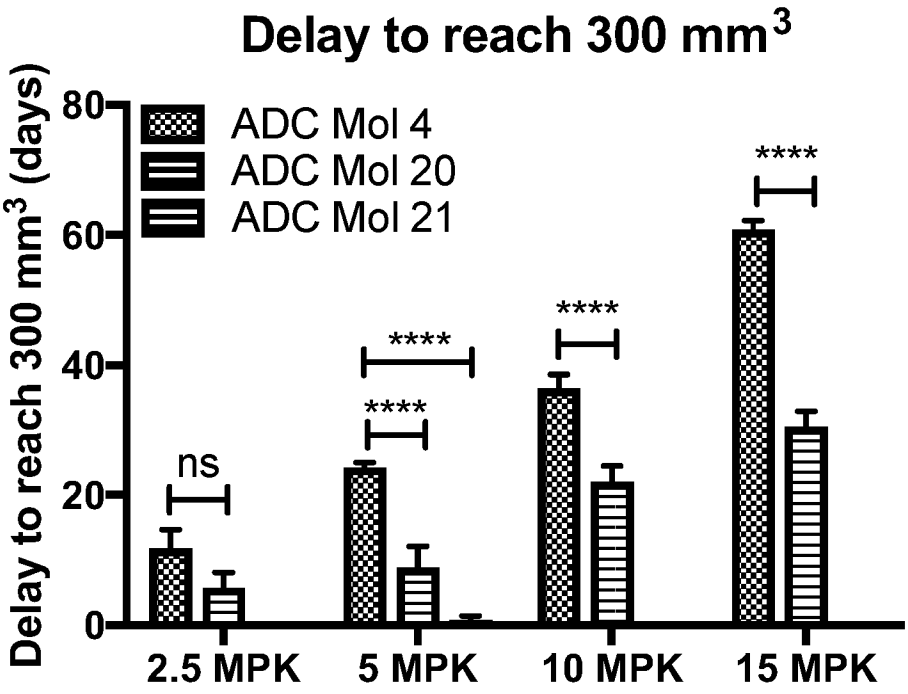


FIG. 19D



ns= not significant

FIG. 20

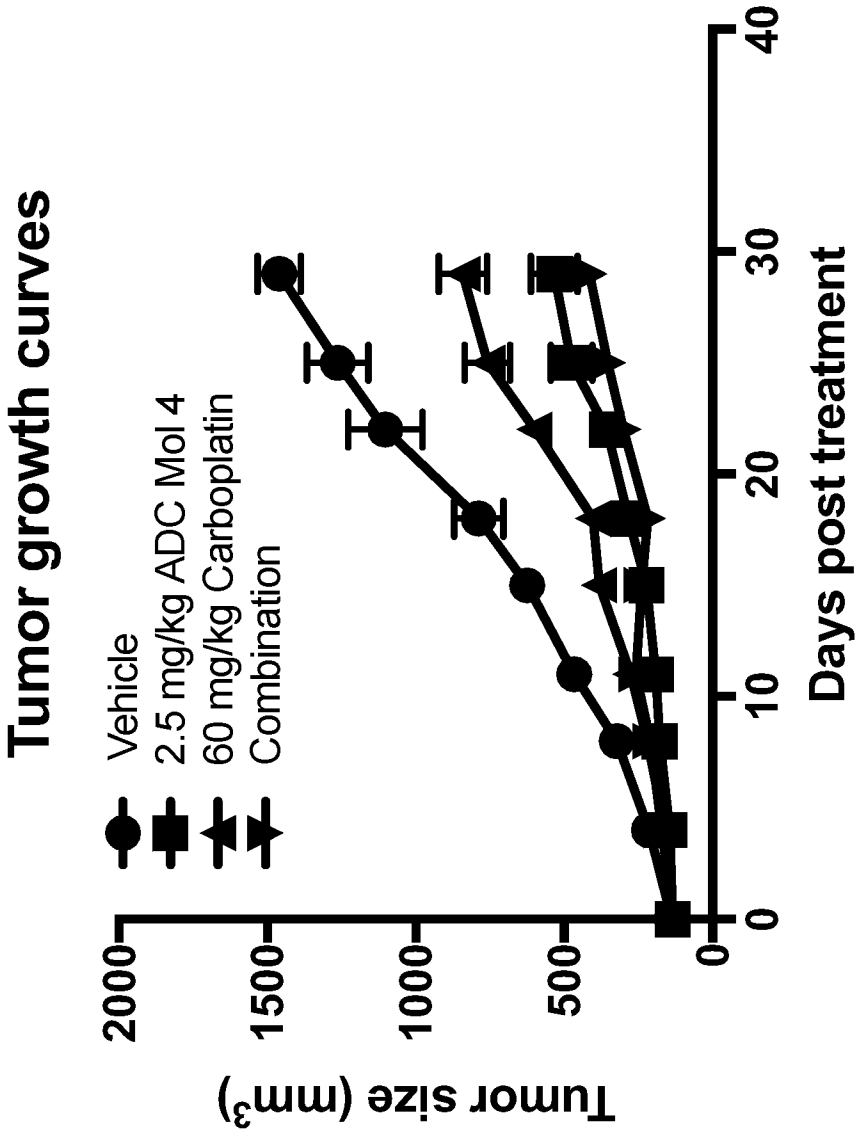
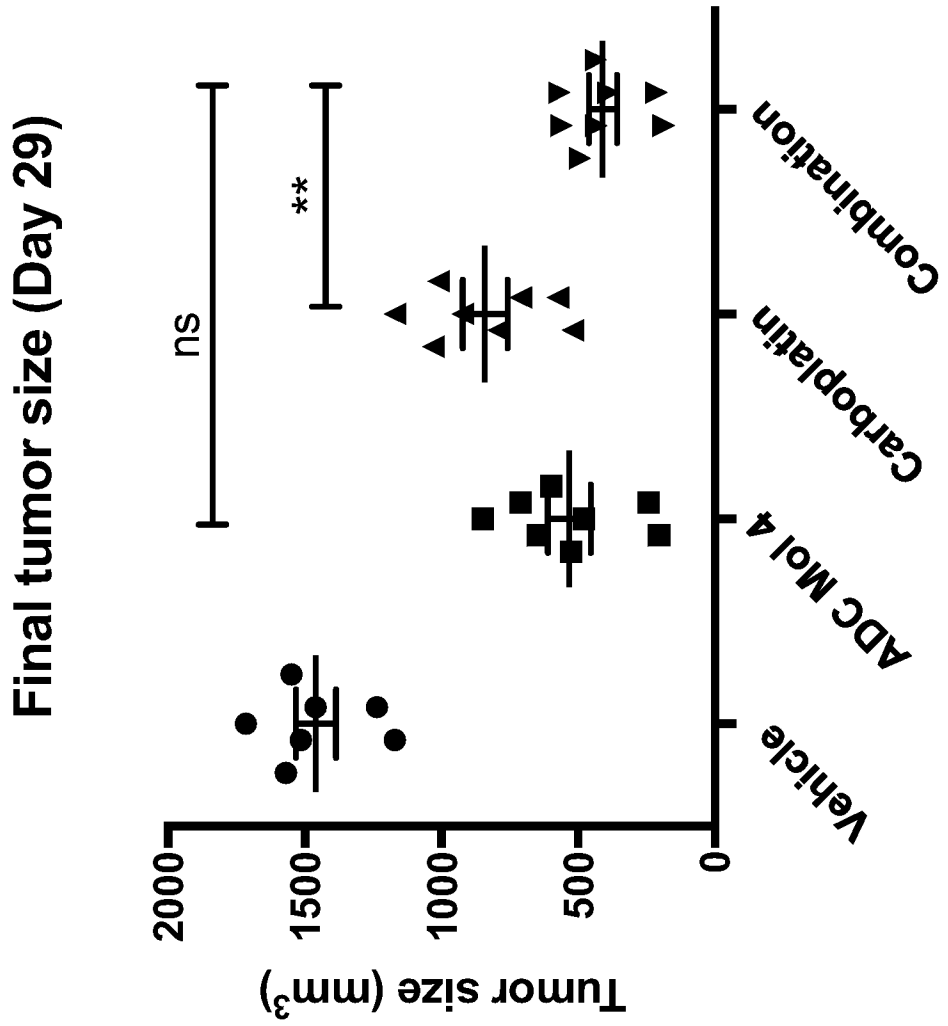
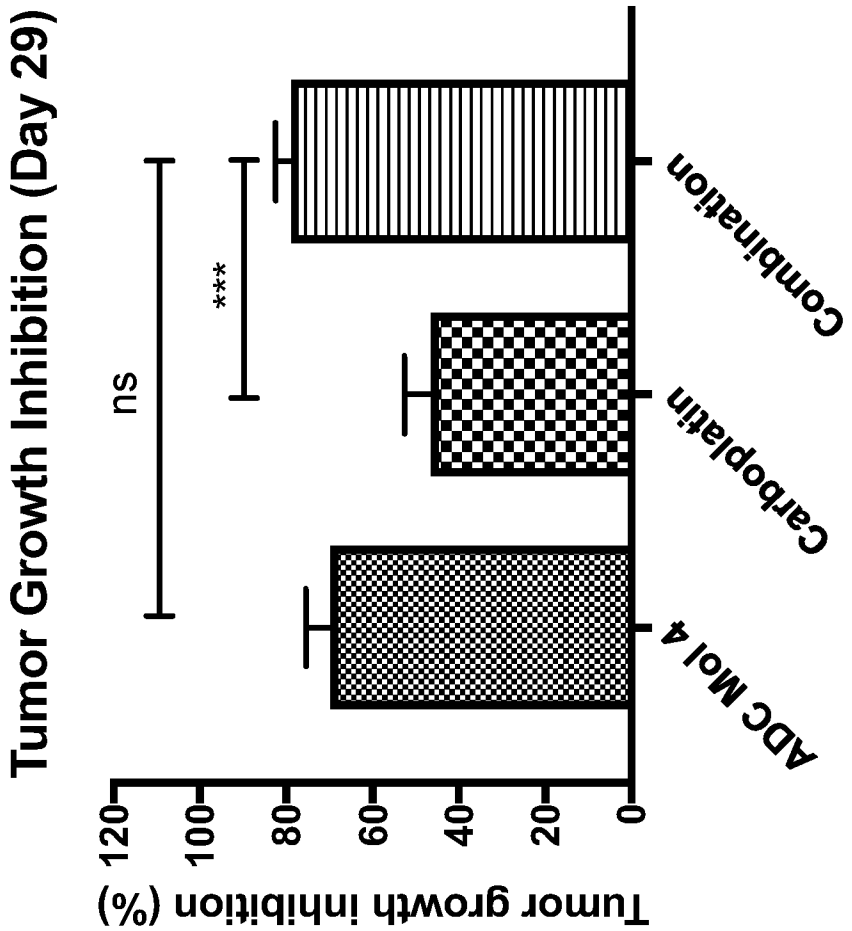

Tumor growth curves
Vehicle
2.5 mg/kg ADC Mol 4
60 mg/kg Carboplatin
Combination
Tumor size (mm³)
2000
1500
1000
500
0
0
10
20
30
40
Days post treatment

FIG. 21A



ns= not significant

FIG. 21B



ns= not significant

FIG. 21C

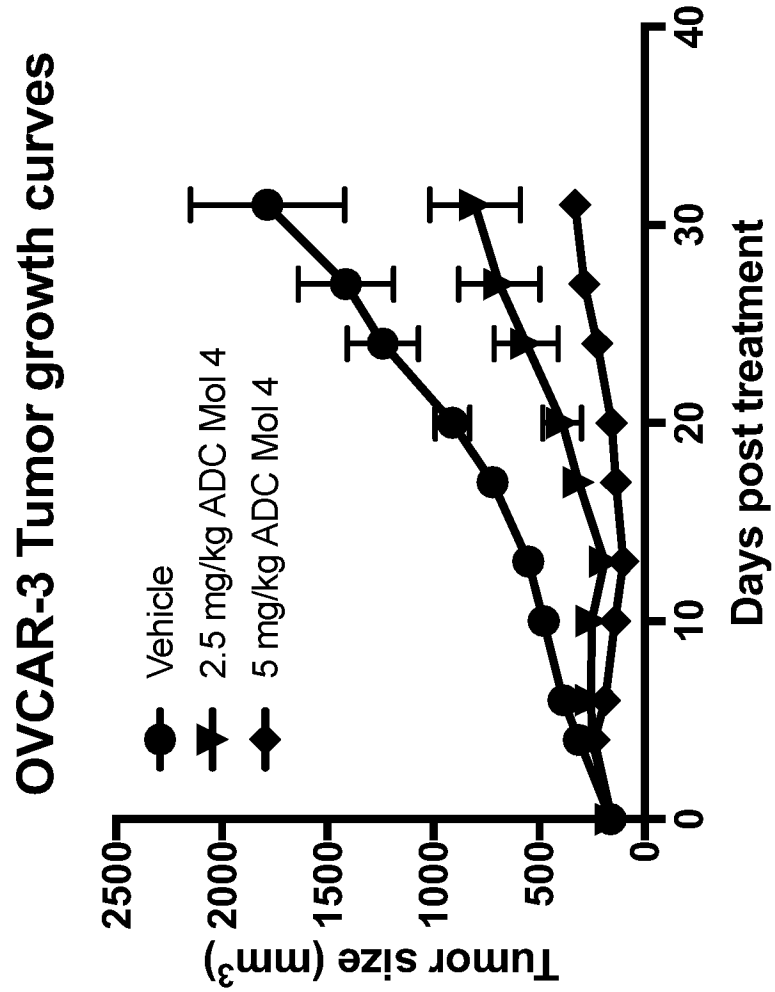

OVCAR-3 Tumor growth curves
Vehicle
2.5 mg/kg ADC Mol 4
5 mg/kg ADC Mol 4
Tumor size (mm3)
2500
2000
1500
1000
500
0
0
10
20
30
40
Days post treatment

FIG. 22A

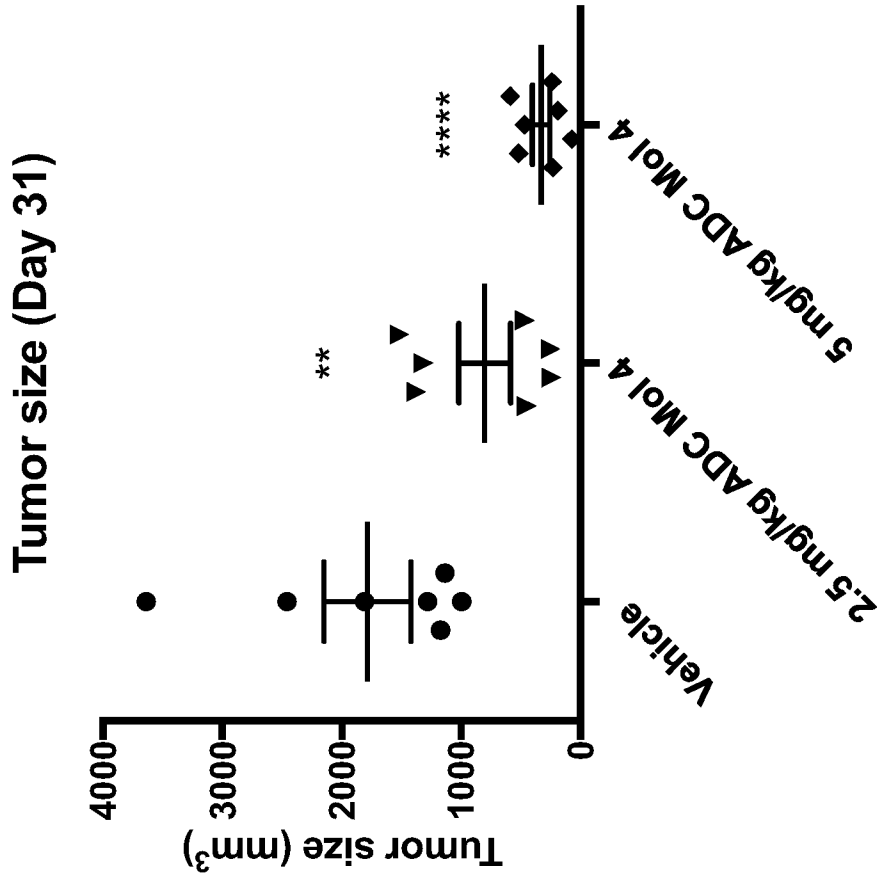

Tumor size (Day 31)
Tumor size (mm3)
4000
3000
2000
1000
0
**
****
Vehicle
2.5 mg/kg ADC Mol 4
5 mg/kg ADC Mol 4

FIG. 22B

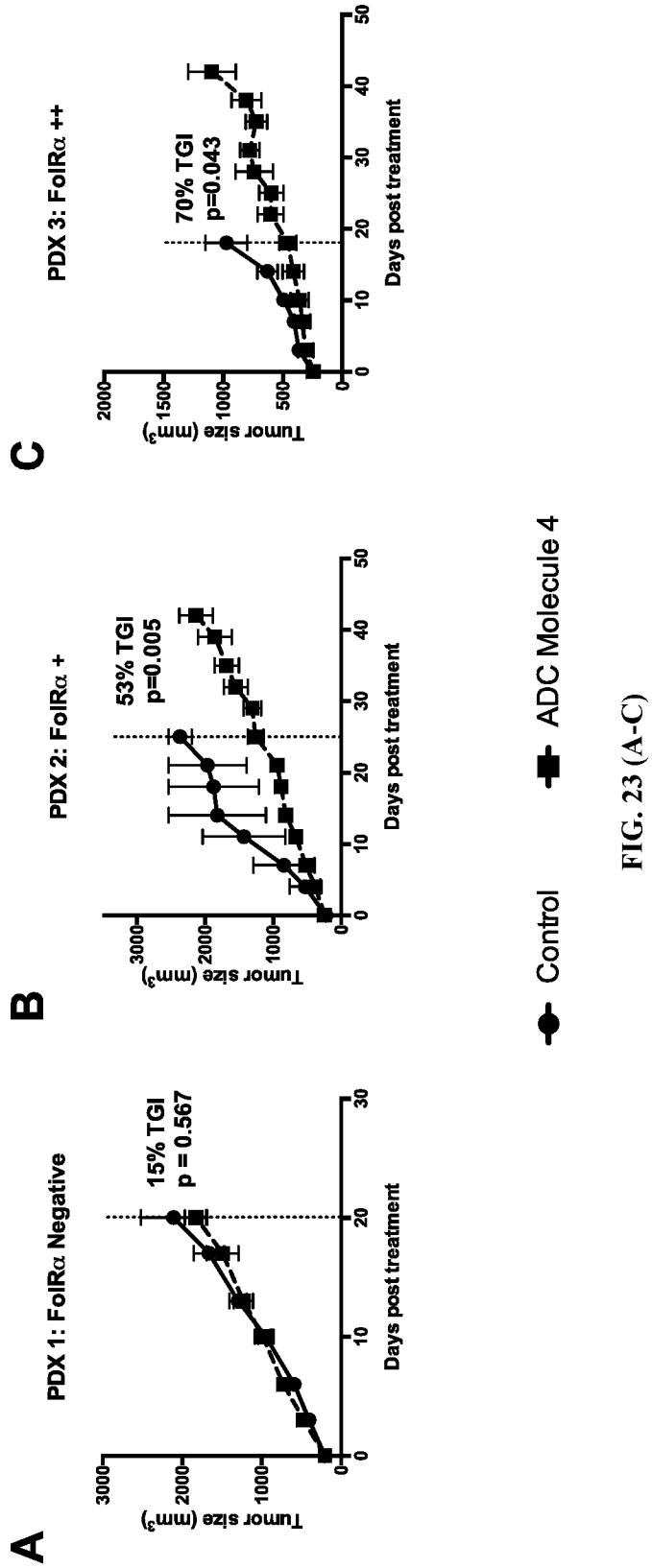

A
PDX 1: FolRα Negative
15% TGI
p = 0.567
Tumor size (mm3)
Days post treatment
B
PDX 2: FolRα +
53% TGI
p=0.005
Tumor size (mm3)
Days post treatment
C
PDX 3: FolRα ++
70% TGI
p=0.043
Tumor size (mm3)
Days post treatment
Control
ADC Molecule 4

FIG. 23 (A-C)

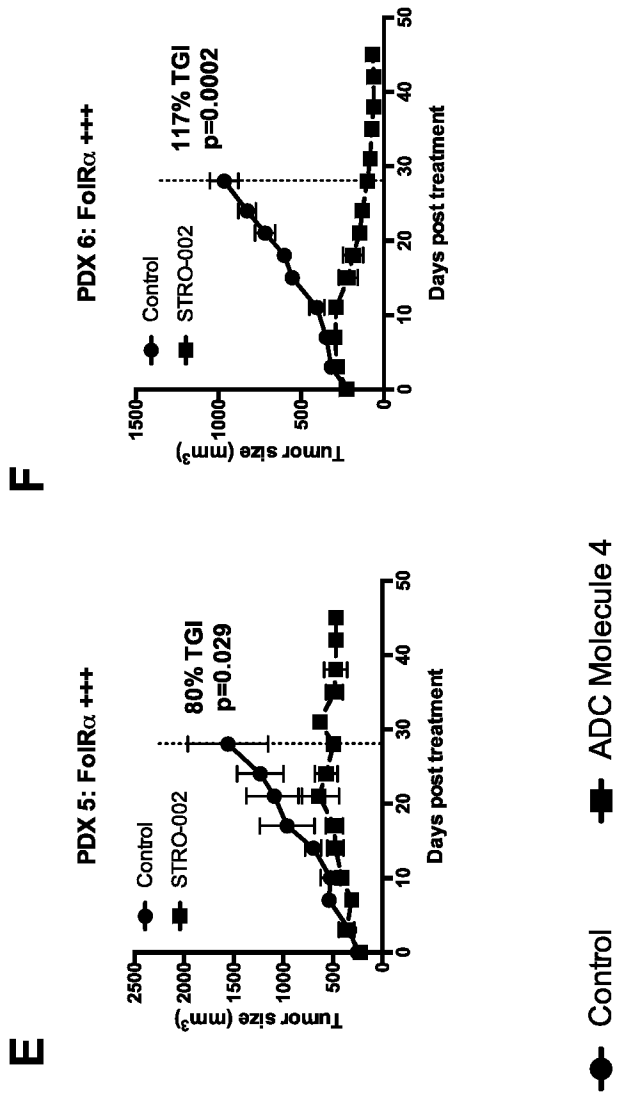

D
PDX 4: FolRα ++
112% TGI
p=0.012
Tumor size (mm3)
Days post treatment
E
PDX 5: FolRα +++
Control
STRO-002
80% TGI
p=0.029
Tumor size (mm3)
Days post treatment
F
PDX 6: FolRα +++
Control
STRO-002
117% TGI
p=0.0002
Tumor size (mm3)
Days post treatment
Control
ADC Molecule 4

FIG. 23 (D-F)

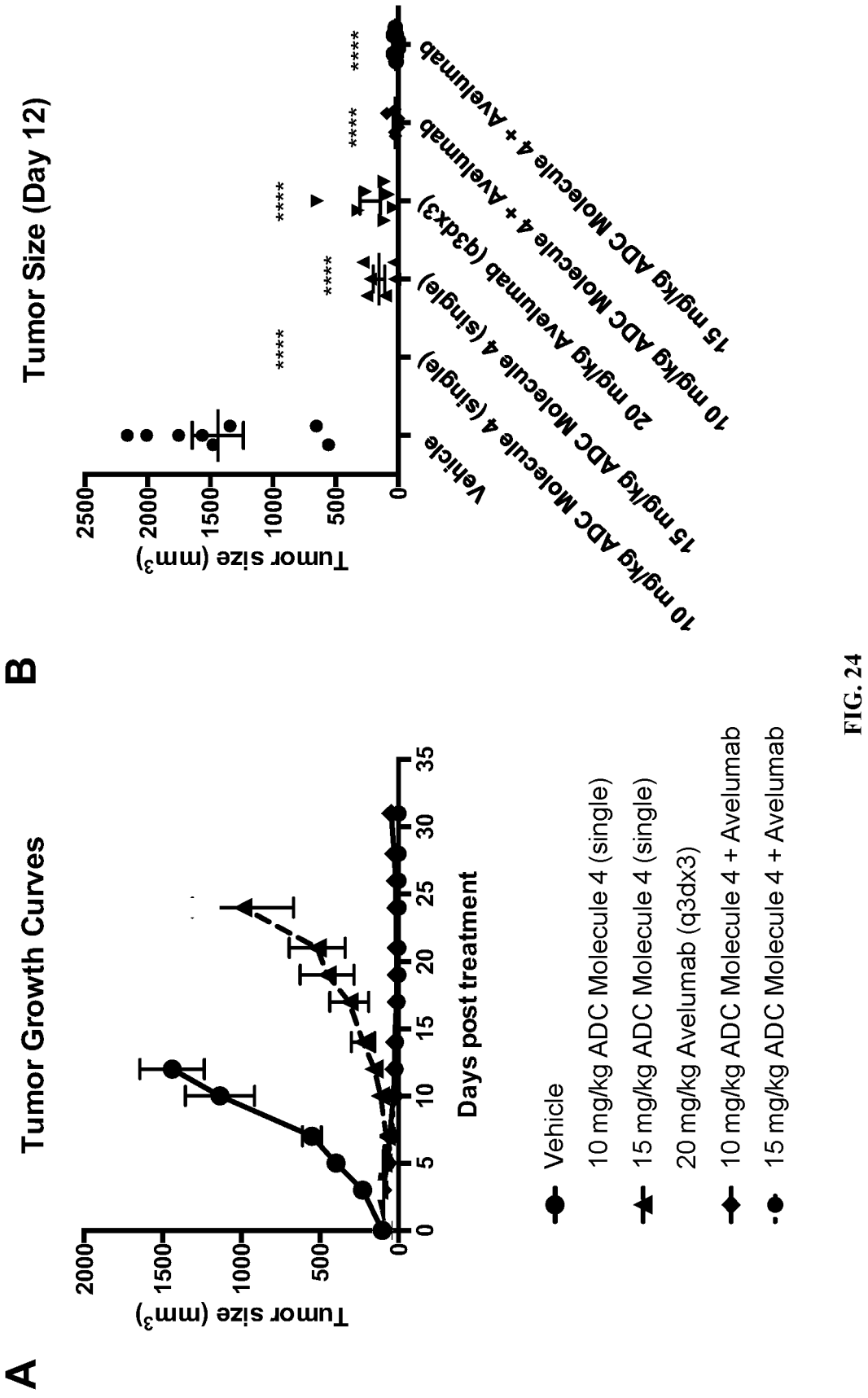

A
Tumor Growth Curves
Tumor size (mm3)
2000
1500
1000
500
0
0
5
10
15
20
25
30
35
Days post treatment
Vehicle
10 mg/kg ADC Molecule 4 (single)
15 mg/kg ADC Molecule 4 (single)
20 mg/kg Avelumab (q3dx3)
10 mg/kg ADC Molecule 4 + Avelumab
15 mg/kg ADC Molecule 4 + Avelumab
B
Tumor Size (Day 12)
Tumor size (mm3)
2500
2000
1500
1000
500
0
****
****
****
****
Vehicle
10 mg/kg ADC Molecule 4 (single)
15 mg/kg ADC Molecule 4 (single)
20 mg/kg Avelumab (q3dx3)
10 mg/kg ADC Molecule 4 + Avelumab
15 mg/kg ADC Molecule 4 + Avelumab

FIG. 24

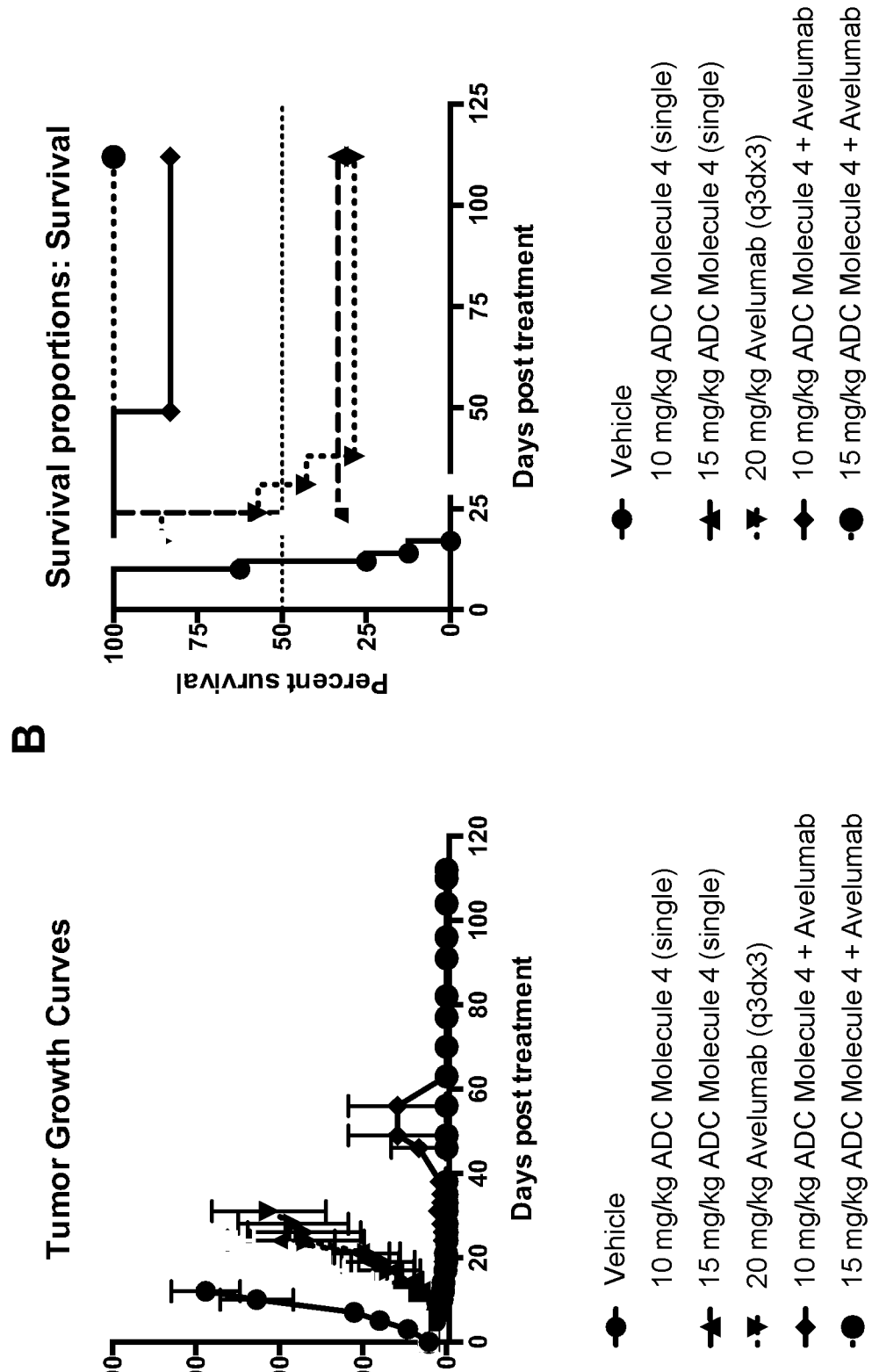

A
Tumor Growth Curves
Tumor size (mm3)
2000
1500
1000
500
0
0
20
40
60
80
100
120
Days post treatment
B
Survival proportions: Survival
Percent survival
100
75
50
25
0
0
25
50
75
100
125
Days post treatment
Vehicle
10 mg/kg ADC Molecule 4 (single)
15 mg/kg ADC Molecule 4 (single)
20 mg/kg Avelumab (q3dx3)
10 mg/kg ADC Molecule 4 + Avelumab
15 mg/kg ADC Molecule 4 + Avelumab

FIG. 25

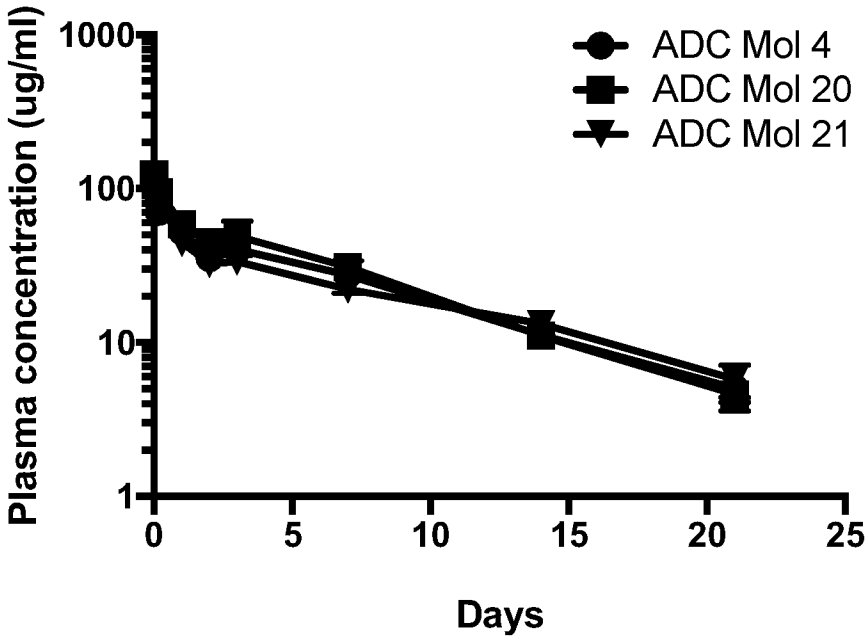

Plasma concentration (ug/ml)
1000
100
10
1
0
5
10
15
20
25
Days
ADC Mol 4
ADC Mol 20
ADC Mol 21

FIG. 26

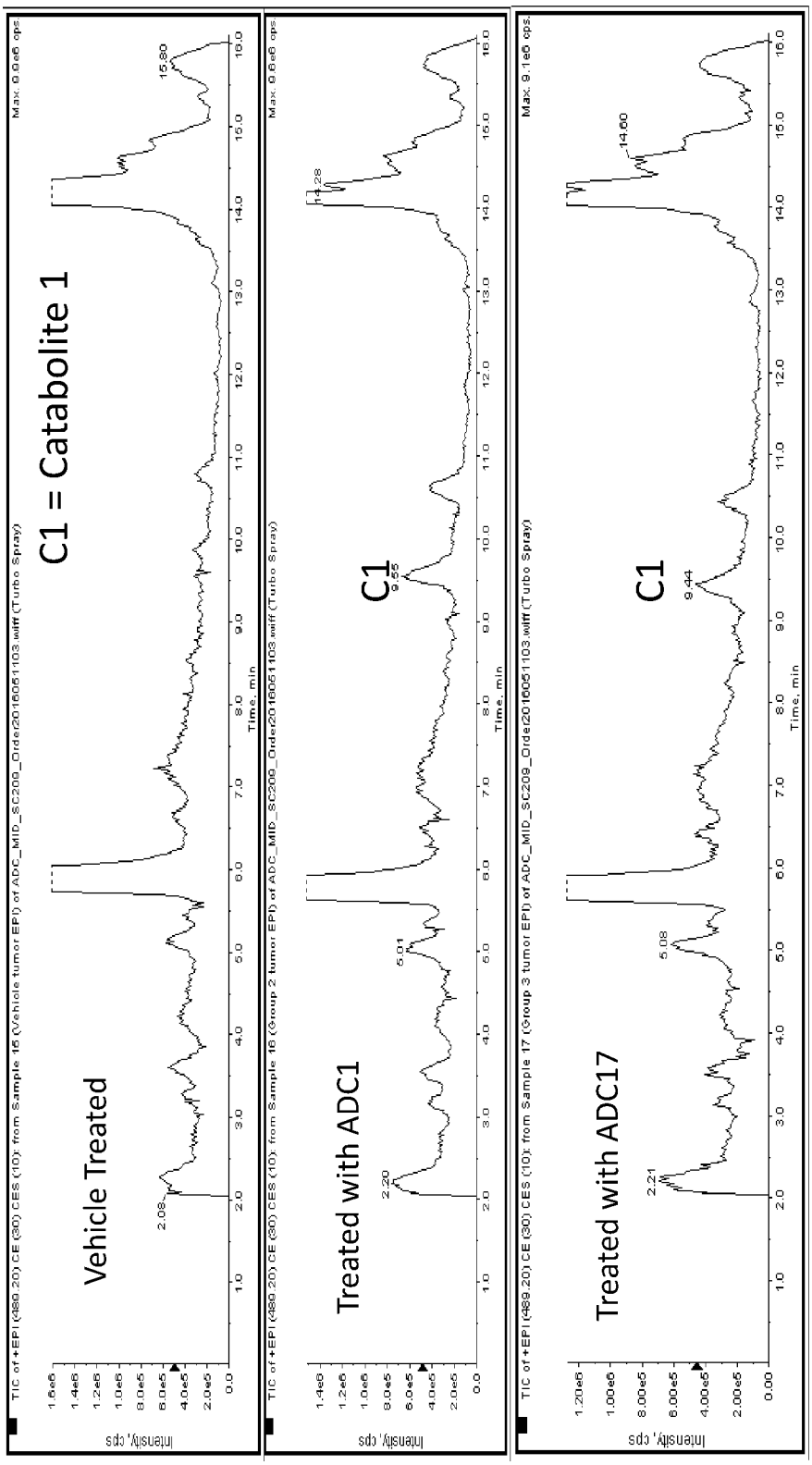

C1 = Catabolite 1
Vehicle Treated
Treated with ADC1
Treated with ADC17
C1
Intensity, cps
Time, min
2.08
15.80
2.20
5.01
9.65
14.28
2.21
5.08
9.44
14.60

FIG. 27

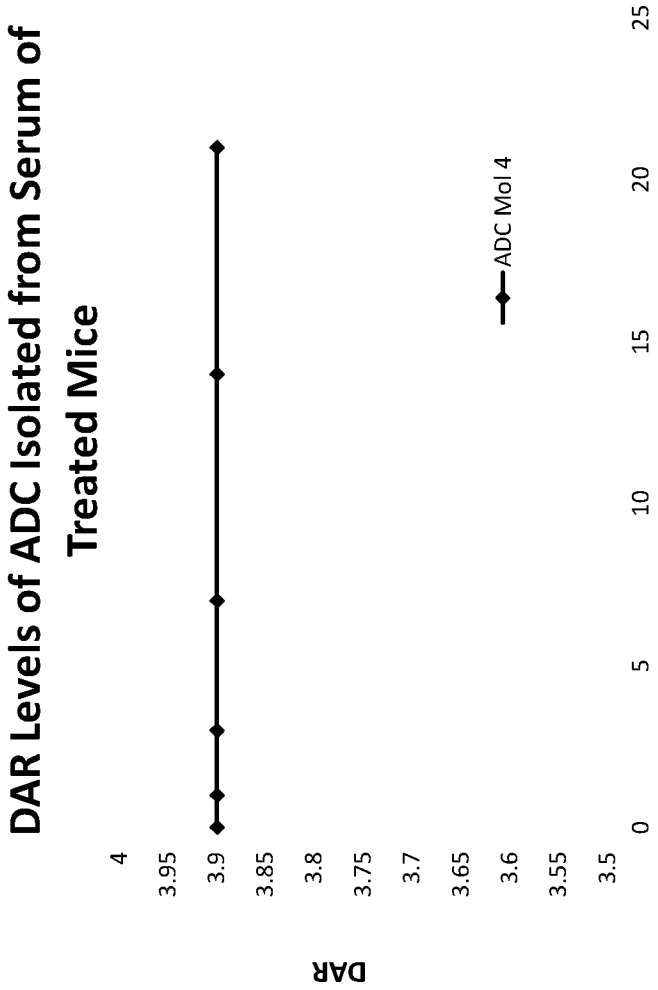
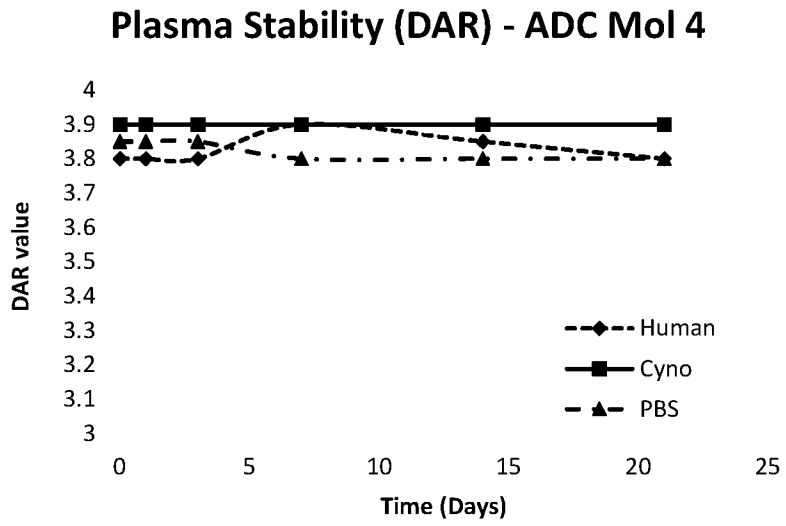

DAR Levels of ADC Isolated from Serum of Treated Mice
DAR
4
3.95
3.9
3.85
3.8
3.75
3.7
3.65
3.6
3.55
3.5
0
5
10
15
20
25
Days
ADC Mol 4

FIG. 28


Plasma Stability (DAR) - ADC Mol 4
DAR value
4
3.9
3.8
3.7
3.6
3.5
3.4
3.3
3.2
3.1
3
0
5
10
15
20
25
Time (Days)
Human
Cyno
PBS

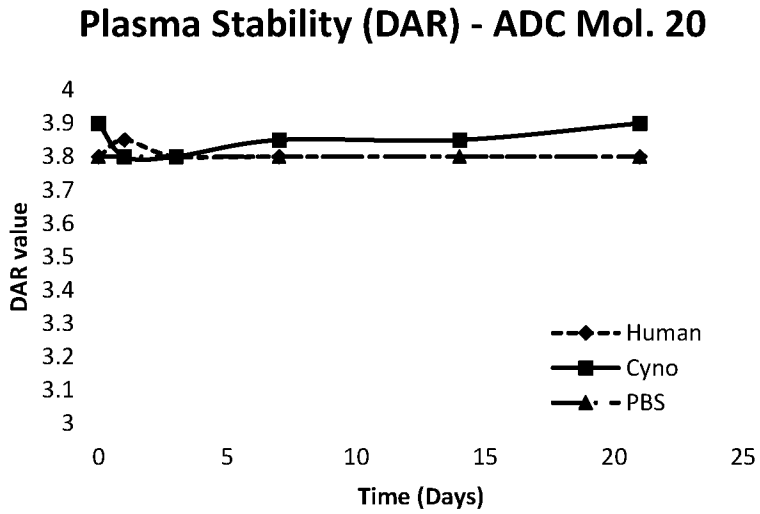

Plasma Stability (DAR) - ADC Mol. 20
DAR value
4
3.9
3.8
3.7
3.6
3.5
3.4
3.3
3.2
3.1
3
0
5
10
15
20
25
Time (Days)
Human
Cyno
PBS

FIG. 29

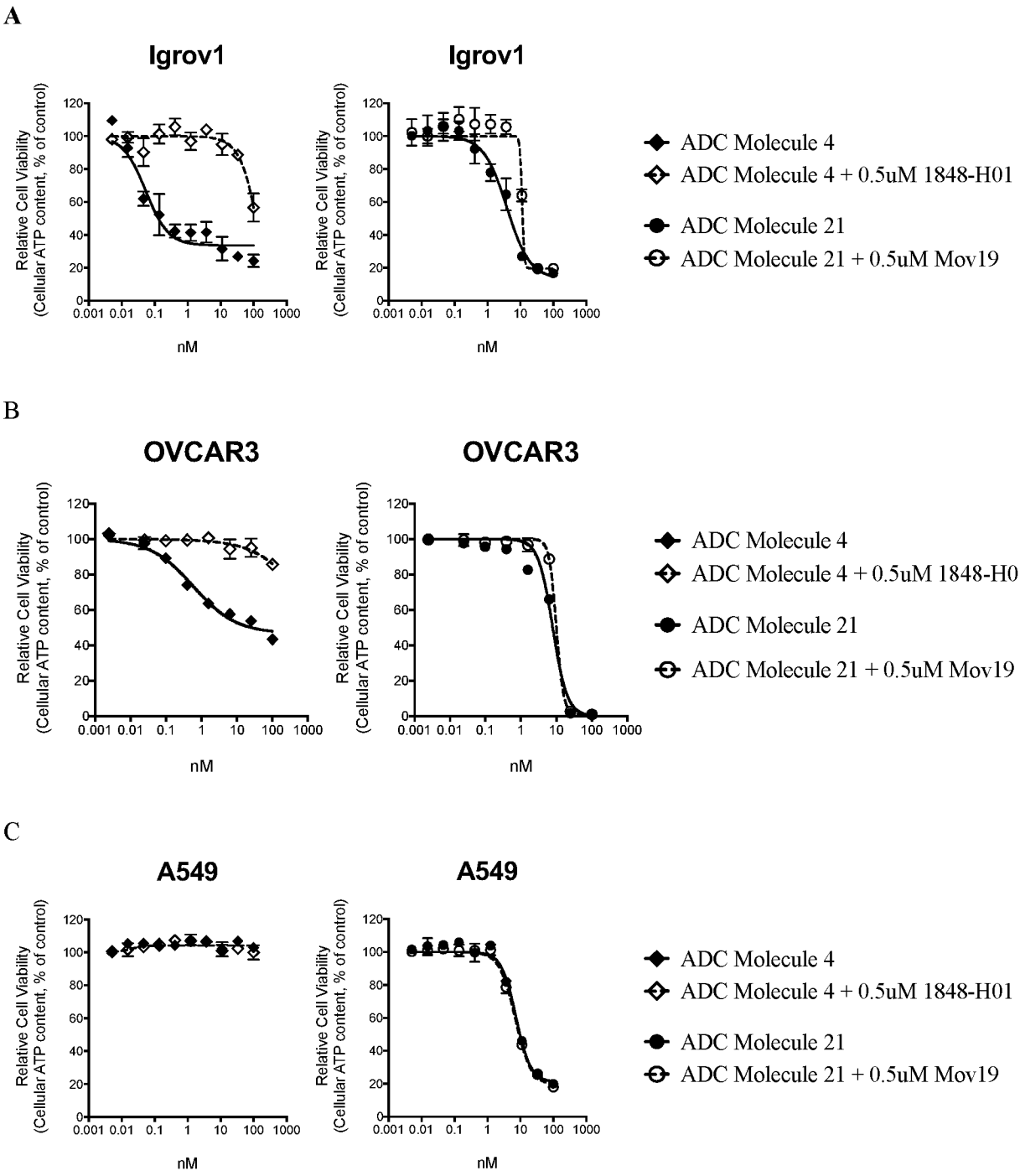

A
Igrov1
Igrov1
Relative Cell Viability (Cellular ATP content, % of control)
nM
ADC Molecule 4
ADC Molecule 4 + 0.5uM 1848-H01
ADC Molecule 21
ADC Molecule 21 + 0.5uM Mov19
B
OVCAR3
OVCAR3
C
A549
A549

FIG. 30

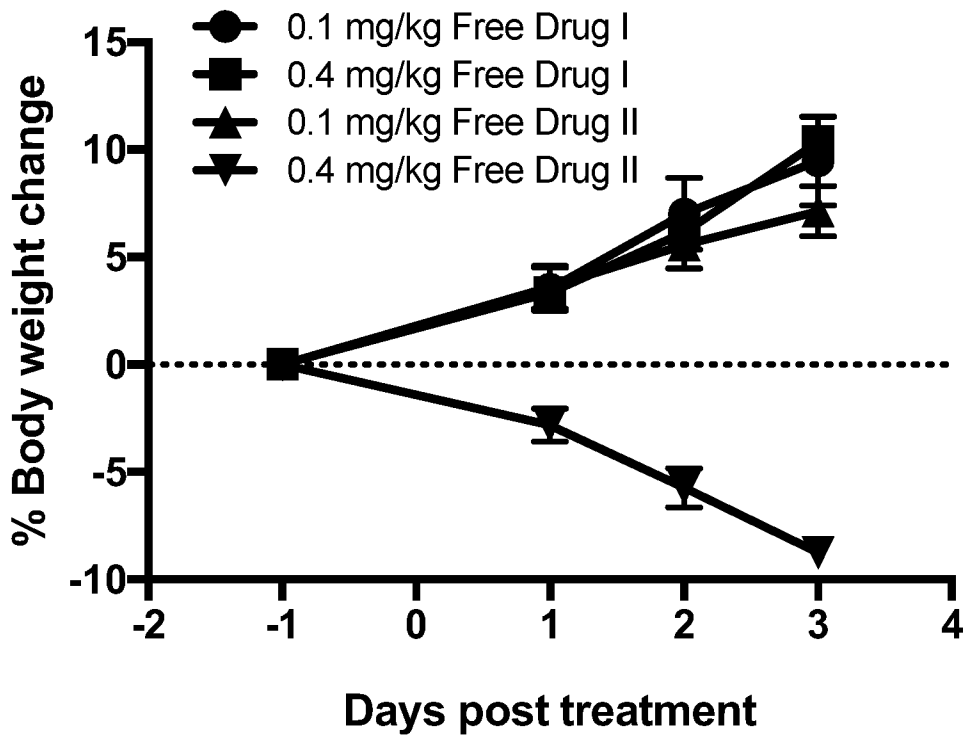

0.1 mg/kg Free Drug I
0.4 mg/kg Free Drug I
0.1 mg/kg Free Drug II
0.4 mg/kg Free Drug II
% Body weight change
15
10
5
0
-5
-10
-2
-1
0
1
2
3
4
Days post treatment

FIG. 31

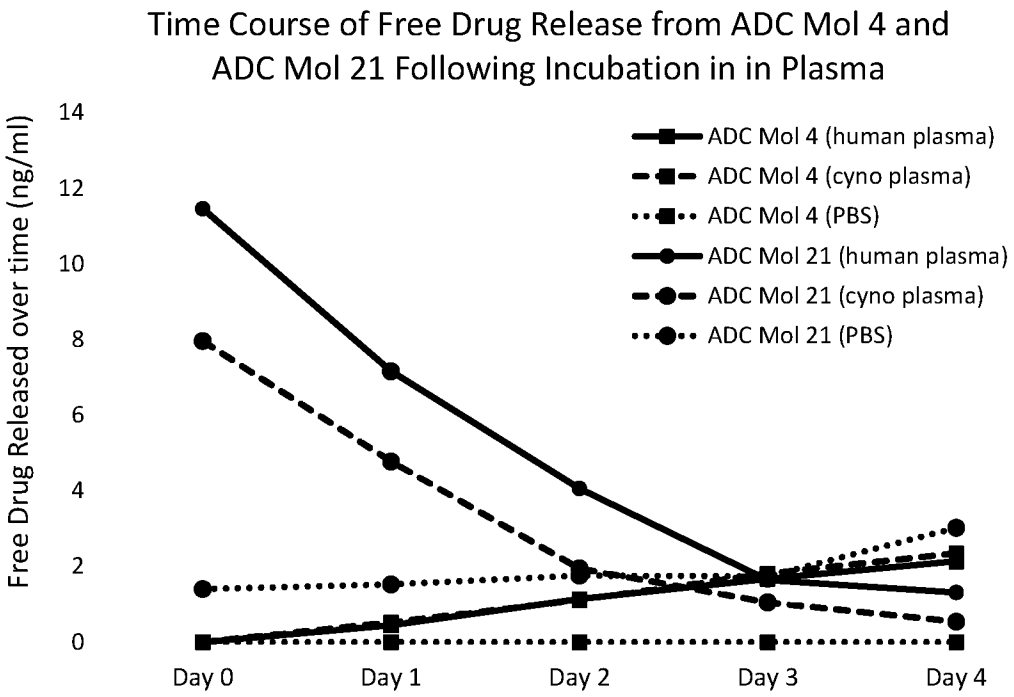

Time Course of Free Drug Release from ADC Mol 4 and ADC Mol 21 Following Incubation in in Plasma
Free Drug Released over time (ng/ml)
14
12
10
8
6
4
2
0
Day 0
Day 1
Day 2
Day 3
Day 4
ADC Mol 4 (human plasma)
ADC Mol 4 (cyno plasma)
ADC Mol 4 (PBS)
ADC Mol 21 (human plasma)
ADC Mol 21 (cyno plasma)
ADC Mol 21 (PBS)

FIG. 32

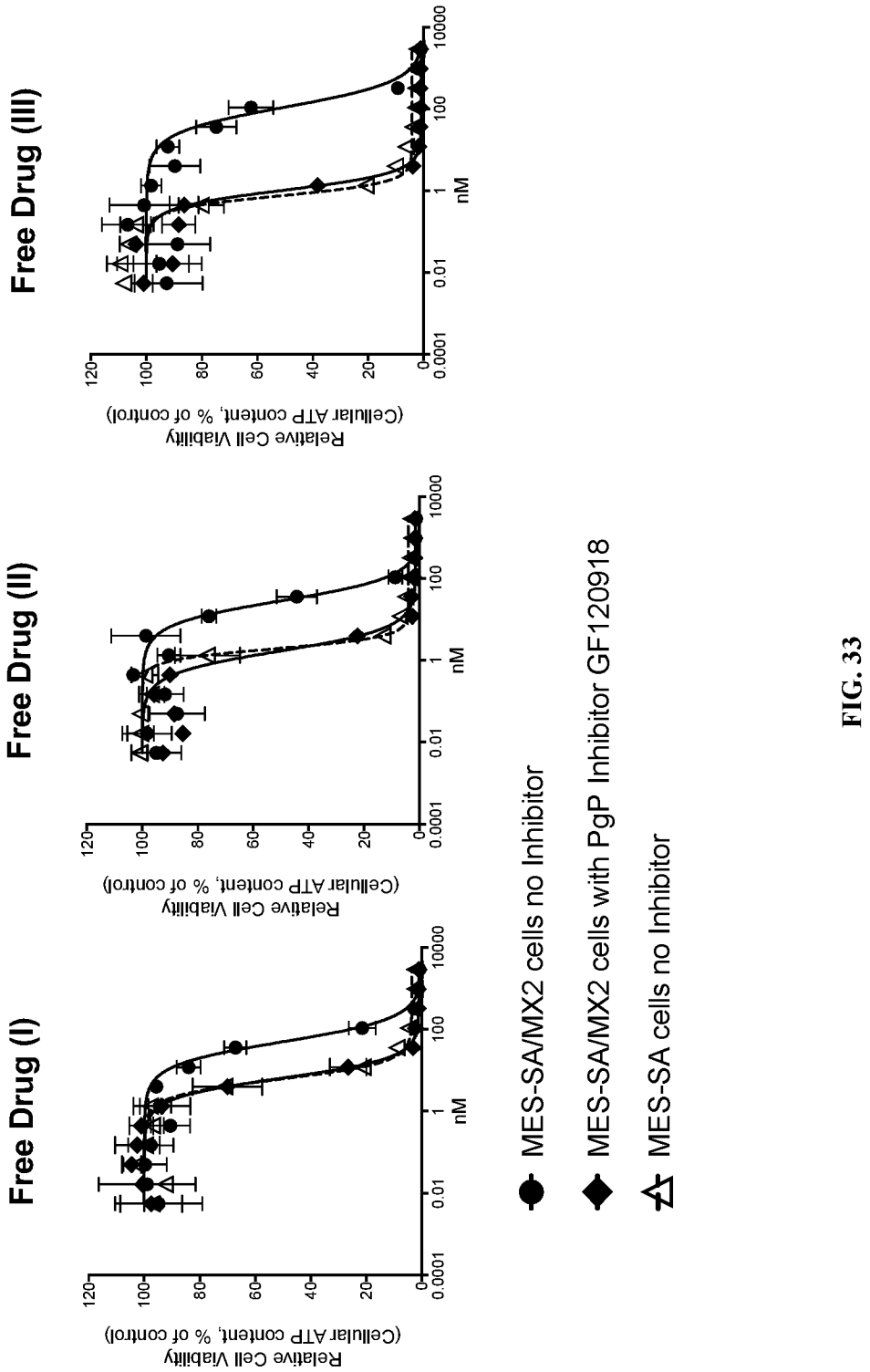
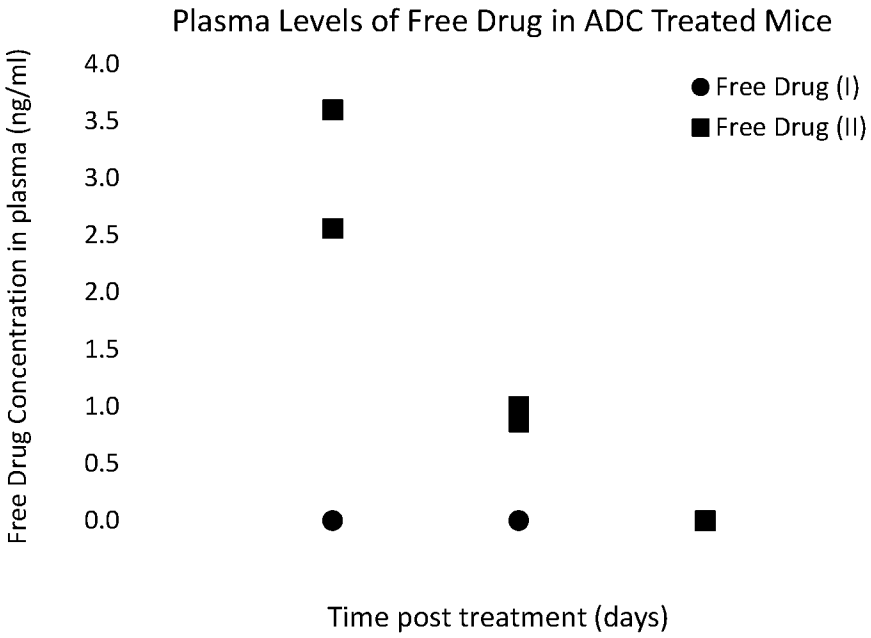

Free Drug (I)
Free Drug (II)
Free Drug (III)
Relative Cell Viability
(Cellular ATP content, % of control)
0
20
40
60
80
100
120
0.0001
0.01
1
100
10000
nM
MES-SA/MX2 cells no Inhibitor
MES-SA/MX2 cells with PgP Inhibitor GF120918
MES-SA cells no Inhibitor

FIG. 33

A


Plasma Levels of Free Drug in ADC Treated Mice
Free Drug Concentration in plasma (ng/ml)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
Free Drug (I)
Free Drug (II)
Time post treatment (days)

B

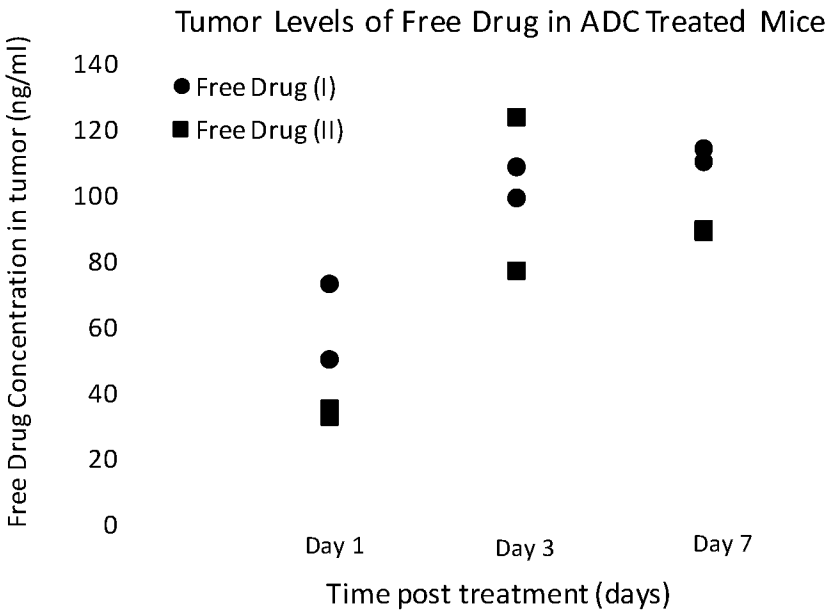

Tumor Levels of Free Drug in ADC Treated Mice
Free Drug Concentration in tumor (ng/ml)
140
120
100
80
60
40
20
0
Free Drug (I)
Free Drug (II)
Day 1
Day 3
Day 7
Time post treatment (days)

FIG. 34

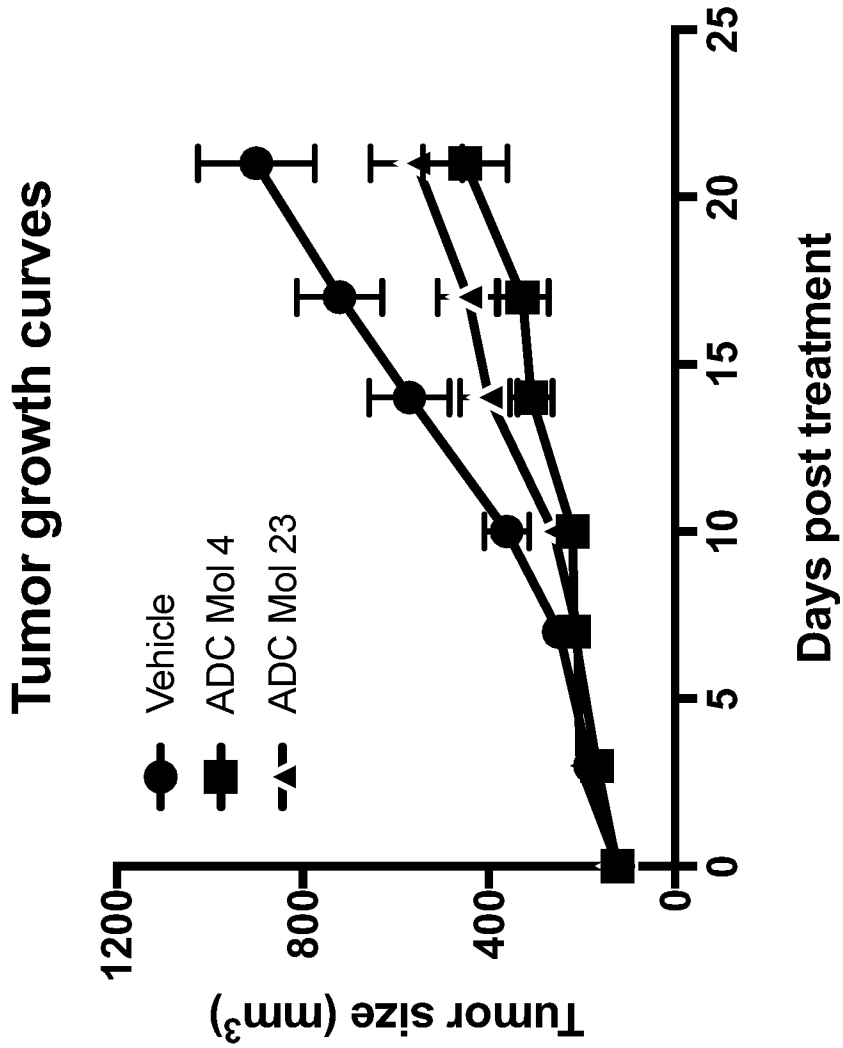

Tumor growth curves
Vehicle
ADC Mol 4
ADC Mol 23
Tumor size (mm3)
1200
800
400
0
0
5
10
15
20
25
Days post treatment

FIG. 35A

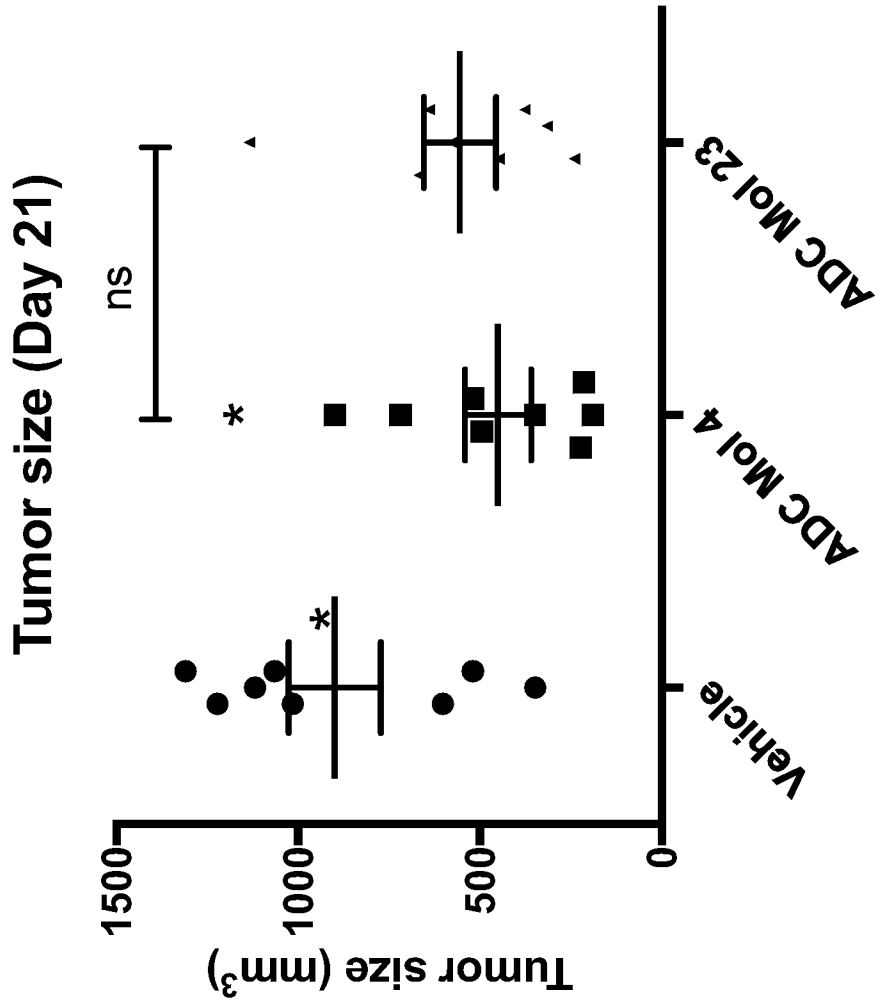

Tumor size (Day 21)
Tumor size (mm3)
1500
1000
500
0
ns
*
*
Vehicle
ADC Mol 4
ADC Mol 23

FIG. 35B

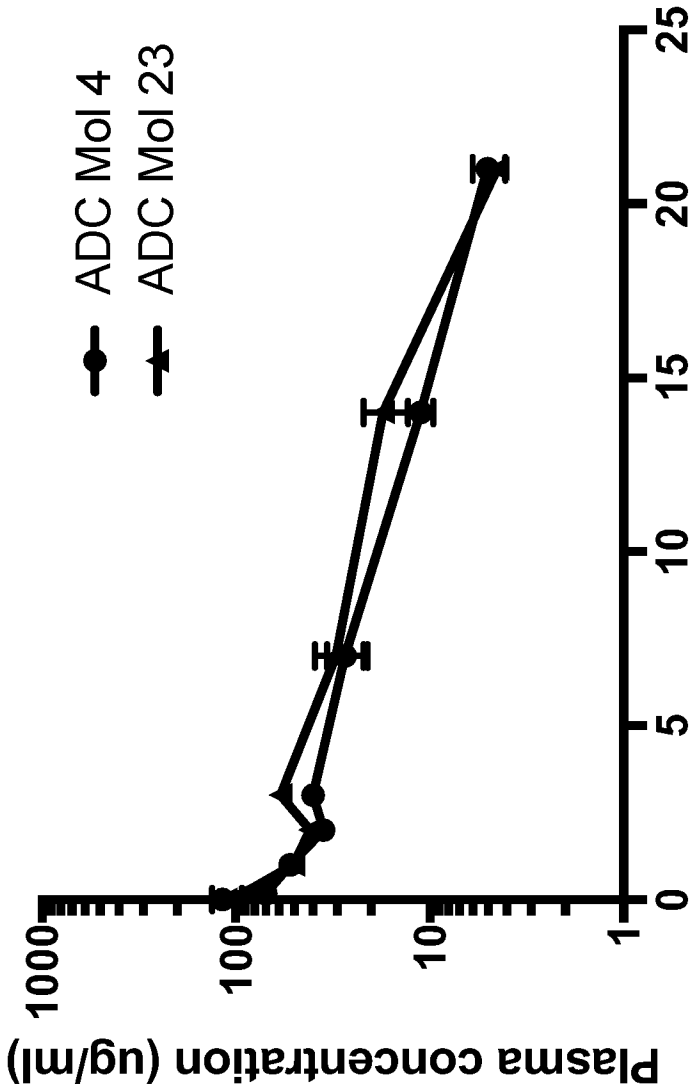

ADC Mol 4
ADC Mol 23
Plasma concentration (ug/ml)
1000
100
10
1
0
5
10
15
20
25
Days

FIG. 36

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2019/051350

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K47/68
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	U.A. MATULONIS ET AL: "Initial safety and activity findings from a phase IB escalation study of mirvetuximab soravtansine, a folate receptor alpha (FR[alpha]-targeting antibody-drug conjugate (ADC), with pembrolizumab in platinum-resistant epithelial ovarian cancer (EOC) patients", GYNECOLOGIC ONCOLOGY., vol. 149, 25 June 2018 (2018-06-25), page 38, XP055664069, GB ISSN: 0090-8258, DOI: 10.1016/j.ygyno.2018.04.085	1-90
Y	whole document	1-90
	----- -/--	

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

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Date of the actual completion of the international search	Date of mailing of the international search report
3 February 2020	10/02/2020

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Birikaki, Lemonia

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Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2019/051350

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	GANG YIN ET AL: "RF1 attenuation enables efficient non-natural amino acid incorporation for production of homogeneous antibody drug conjugates", SCIENTIFIC REPORTS, vol. 7, no. 1, 8 June 2017 (2017-06-08), XP055527126, DOI: 10.1038/s41598-017-03192-z whole document and in particular the abstract and discussion ----	1-90
Y	WO 2017/132617 A1 (SUTRO BIOPHARMA INC [US]) 3 August 2017 (2017-08-03) claims 1-40 ----	1-90
X,P	WO 2019/055931 A1 (SUTRO BIOPHARMA INC [US]) 21 March 2019 (2019-03-21) paragraph [00363] example 24 claims 1-54 ----	1-90
X,P	WO 2019/055909 A1 (SUTRO BIOPHARMA INC [US]) 21 March 2019 (2019-03-21) example 24 claims 1-54 paragraph [00528] ----	1-90
X,P	WO 2018/213260 A1 (IMMUNOGEN INC [US]; MERCK SHARP & DOHME [US]) 22 November 2018 (2018-11-22) claims 1-67 paragraph [0099] ----	1-90

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2019/051350

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