ANTI-CD74 ANTIBODY CONJUGATES, COMPOSITIONS COMPRISING ANTI-CD74 ANTIBODY CONJUGATES AND METHODS OF USING ANTI-CD74 ANTIBODY CONJUGATES

FIELD

[0001] Provided herein are antibody conjugates with binding specificity for CD74 and compositions comprising the antibody conjugates, including pharmaceutical compositions, methods of producing the conjugates, and methods of using the conjugates and compositions for therapy. The conjugates and 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 conjugates and compositions are also useful in methods of treatment, prevention, detection, and diagnosis of autoimmune diseases, infectious diseases, and inflammatory conditions.

BACKGROUND

[0002] Human leukocyte antigen (HLA) class II histocompatibility antigen gamma chain (also known as HLA-DR antigens-associated invariant chain or CD74 (Cluster of Differentiation 74)) is a protein that is involved in the formation and transport of major histocompatibility complex (MHC) class II protein. *See* Claesson *et al*., *Proc. Natl. Acad. Sci. U.S.A.*, 1983, 80:7395-7399; Kudo *et al*., *Nucleic Acids Res.*, 1985, 13:8827-8841; and Cresswell, *Ann. Rev. Immunol.*, 1994, 12:259-291.

[0003] One function of CD74 is to regulate peptide loading onto MHC class II heterodimers in intracellular compartments, to prevent MHC class II from binding cellular peptides. The full range of functionality of cell surface-expressed CD74 is not yet known. However, studies have demonstrated that CD74 is a receptor for the pro-inflammatory cytokine macrophage migration inhibitory factor (MIF). Binding of MIF to CD74 activates downstream signaling through the MAPK and Akt pathways and promotes cell proliferation and survival. *See* Gore *et al*., *J. Biol. Chem.*, 2008, 283:2784-2792; and Starlets *et al*., *Blood*, 2006, 107:4807-4816.

[0004] Upregulation of CD74 expression has been observed in cancers and autoimmune disease (Borghese *et al*., *Exp. Op. Ther. Targets*, 2011, 15:237-251), as well as in infection (Hofman *et al*., *Modern Pathology*, 2007, 20:974-989) and inflammatory conditions (Vera *et al*., *Exp. Biol. & Med.*, 2008, 233:620-626). CD74 is known to be expressed at moderate to high levels on a variety of hematological tumors including B-cell lymphoma, leukemia, and multiple myeloma. Burton *et al*., *Clin. Cancer Res.*, 2004,

10:6606-6611. CD74 expression is also known to be a key factor associated with the progression of pancreatic cancer. Zhang *et al*., *Hepatobiliary Pancreat. Dis. Int.*, 2014, 13:81-86.

[0005] In view of the role of CD74 in multiple disease processes, there is a need for improved methods of modulating the interaction of CD74 with its ligands and the downstream signaling processes activated by CD74. Moreover, given the upregulation of CD74 in several diseases, there is also a need for therapeutics that specifically target cells and tissues overexpressing CD74. Antibody conjugates to CD74 could be used to deliver therapeutic or diagnostic payload moieties to target cells expressing CD74 for the treatment or diagnosis of such diseases.

SUMMARY

[0006] Provided herein are antibody conjugates that selectively bind CD74. The antibody conjugates comprise an antibody that binds CD74 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. CD74 antibodies are described in detail herein, as are useful payload moieties, and useful linkers.

[0007] Also provided are compositions comprising the antibody conjugates. 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.Further provided herein are kits comprising the antibody conjugates or pharmaceutical compositions.

[0008] Also provided herein are methods of using the anti-CD74 antibody conjugates. In some embodiments, the methods are methods of delivering one or more payload moieties to a target cell or tissue expressing CD74. 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 antibody conjugates are used to treat a disease or condition. In some embodiments, the disease or condition is selected from a cancer, an autoimmune disease, an infectious disease, or an inflammatory condition. In some embodiments, the disease or condition is a B-cell lymphoma. In some embodiments, the disease or condition is non-Hodgkin’s lymphoma. In some embodiments, the disease or condition is leukemia. In some embodiments, the disease or condition is pancreatic cancer. In some embodiments, the disease or condition is multiple myeloma.

[0008a] According to an aspect, there is provided an antibody conjugate comprising an antibody that specifically binds to CD74 linked site-specifically to at least one payload moiety, wherein the antibody comprises a non-natural amino acid at a site 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 wherein the antibody further comprises:

a. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 1 and 33; a CDR-H2 comprising at least one of SEQ ID NOs: 65 and 97; and a CDR-H3 comprising SEQ ID NO: 129;

b. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 2 and 34; a CDR-H2 comprising at least one of SEQ ID NOs: 66 and 98; and a CDR-H3 comprising SEQ ID NO: 130;

c. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 3 and 35; a CDR-H2 comprising at least one of SEQ ID NOs: 67 and 99; and a CDR-H3 comprising SEQ ID NO: 131;

d. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 4 and 36; a CDR-H2 comprising at least one of SEQ ID NOs: 68 and 100; and a CDR-H3 comprising SEQ ID NO: 132;

e. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 5 and 37; a CDR-H2 comprising at least one of SEQ ID NOs: 69 and 101; and a CDR-H3 comprising SEQ ID NO: 133;

f. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 6 and 38; a CDR-H2 comprising at least one of SEQ ID NOs: 70 and 102; and a CDR-H3 comprising SEQ ID NO: 134;

g. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 7 and 39; a CDR-H2 comprising at least one of SEQ ID NOs: 71 and 103; and a CDR-H3 comprising SEQ ID NO: 135;

h. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 8 and 40; a CDR-H2 comprising at least one of SEQ ID NOs: 72 and 104; and a CDR-H3 comprising SEQ ID NO: 136;

i. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 9 and 41; a CDR-H2 comprising at least one of SEQ ID NOs: 73 and 105; and a CDR-H3 comprising SEQ ID NO: 137;

j. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 10 and 42; a CDR-H2 comprising at least one of SEQ ID NOs: 74 and 106; and a CDR-H3 comprising SEQ ID NO: 138;

k. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 11 and 43; a CDR-H2 comprising at least one of SEQ ID NOs: 75 and 107; and a CDR-H3 comprising SEQ ID NO: 139;

l. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 12 and 44; a CDR-H2 comprising at least one of SEQ ID NOs: 76 and 108; and a CDR-H3 comprising SEQ ID NO: 140;

m. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 13 and 45; a CDR-H2 comprising at least one of SEQ ID NOs: 77 and 109; and a CDR-H3 comprising SEQ ID NO: 141;

n. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 14 and 46; a CDR-H2 comprising at least one of SEQ ID NOs: 78 and 110; and a CDR-H3 comprising SEQ ID NO: 142;

o. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 15 and 47; a CDR-H2 comprising at least one of SEQ ID NOs: 79 and 111; and a CDR-H3 comprising SEQ ID NO: 143;

p. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 16 and 48; a CDR-H2 comprising at least one of SEQ ID NOs: 80 and 112; and a CDR-H3 comprising SEQ ID NO: 144;

q. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 17 and 49; a CDR-H2 comprising at least one of SEQ ID NOs: 81 and 113; and a CDR-H3 comprising SEQ ID NO: 145;

r. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 18 and 50; a CDR-H2 comprising at least one of SEQ ID NOs: 82 and 114; and a CDR-H3 comprising SEQ ID NO: 146;

s. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 19 and 51; a CDR-H2 comprising at least one of SEQ ID NOs: 83 and 115; and a CDR-H3 comprising SEQ ID NO: 147;

t. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 20 and 52; a CDR-H2 comprising at least one of SEQ ID NOs: 84 and 116; and a CDR-H3 comprising SEQ ID NO: 148;

u. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 21 and 53; a CDR-H2 comprising at least one of SEQ ID NOs: 85 and 117; and a CDR-H3 comprising SEQ ID NO: 149;

v. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 22 and 54; a CDR-H2 comprising at least one of SEQ ID NOs: 86 and 118; and a CDR-H3 comprising SEQ ID NO: 150;

w. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 23 and 55; a CDR-H2 comprising at least one of SEQ ID NOs: 87 and 119; and a CDR-H3 comprising SEQ ID NO: 151;

x. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 24 and 56; a CDR-H2 comprising at least one of SEQ ID NOs: 88 and 120; and a CDR-H3 comprising SEQ ID NO: 152;

y. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 25 and 57; a CDR-H2 comprising at least one of SEQ ID NOs: 89 and 121; and a CDR-H3 comprising SEQ ID NO: 153;

z. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 26 and 58; a CDR-H2 comprising at least one of SEQ ID NOs: 90 and 122; and a CDR-H3 comprising SEQ ID NO: 154;

aa. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 27 and 59; a CDR-H2 comprising at least one of SEQ ID NOs: 91 and 123; and a CDR-H3 comprising SEQ ID NO: 155; or

bb. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 28 and 60; a CDR-H2 comprising at least one of SEQ ID NOs: 92 and 124; and a CDR-H3 comprising SEQ ID NO: 156.

[0008b] According to another aspect there is provided a kit comprising an antibody conjugate described herein, and instructions for use of the antibody conjugate.

[0008c] According to another aspect there is provided a pharmaceutical composition comprising the antibody conjugate described herein and a pharmaceutically acceptable carrier.

[0008d] According to another aspect there is provided an antibody conjugate described herein, or a pharmaceutical composition described herein for use in treating or preventing a disease or condition in a subject

[0008e] According to another aspect there is provided an antibody conjugate described herein, or a pharmaceutical composition described herein for use in diagnosing a disease or condition in a subject.

BRIEF DESCRIPTION OF THE DRAWINGS

[0009] **FIG. 1** provides a comparison of the Kabat and Chothia numbering systems for CDR-H1. *See* 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.

[0010] **FIG. 2A** provides protein yield of antibodies expressed according to an example herein; and **FIG. 2B** provides SDS-PAGE analysis of the antibodies under non-reducing and reducing conditions.

[0011] **FIG. 3A** is a plot illustrating body weight change (BWC) as a function of time in a disseminated ARP-1 multiple myeloma model after administration of an anti-CD74 antibody-drug conjugate as disclosed herein. **FIG. 3B** provides a scatter plot of the individual body weight data for each experimental group on day 46 of post-tumor cell inoculation.

[0012] **FIG. 4A** includes images of resulting tumor masses formed in and around ovaries and kidneys from a disseminated ARP-1 multiple myeloma model after administration of an anti-CD74 antibody-drug conjugate as disclosed herein. **FIG. 4B** provides a bar graph of the final tumor tissue weight for each experimental group.

[0013] **FIG. 5A** provides representative flow cytometry dot plots for percentage of human CD138 positive ARP-1 myeloma cells present in the bone marrow (femur and tibia) after administration of an anti-CD74 antibody-drug conjugate as disclosed herein. **FIG. 5B** provides the flow cytometry data as a bar graph of the percentage of human CD138 positive ARP-1 myeloma cells for each experimental group.

[0014] **FIG. 6A** shows body weight change (BWC) as a function of time in a disseminated MM.1S multiple myeloma model after administration of an anti-CD74 antibody-drug conjugate as disclosed herein. **FIG. 6B** provides a Kaplan-Meier survival plot (right) in the multiple myeloma model for each treatment group. **FIG 6C** illustrates bar charts of the percentage of human CD138 positive MM.1S myeloma cells present in the bone marrow on day 32 post-tumor inoculation for each experimental group after administration of an anti-CD74 antibody-drug conjugate as disclosed herein. **FIG 6D** illustrates bar charts of the percentage of human CD138 positive MM.1S myeloma cells present in the bone marrow on day 129 post-tumor inoculation for the experimental groups after administration of anti-CD74 antibody-drug conjugates as disclosed herein.

[0015] **FIG. 7A** provides a tumor growth curve as a function of time in a non-Hodgkin's lymphoma model after administration of an anti-CD74 antibody-drug conjugate as

disclosed herein. **FIG. 7B** provides a scatter plot of individual tumor size on day 18 for each experimental group in the non-Hodgkin's lymphoma model after administration of the anti-CD74 antibody-drug conjugate.

[0016] **FIG. 8** provides a plot of body weight change (BWC) as a function of time in a non-Hodgkin's lymphoma animal model after administration of anti-CD74 antibody-drug conjugates as disclosed herein.

DETAILED DESCRIPTION

1. Definitions

[0017] 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/or parameters unless otherwise noted.

[0018] As used herein, the singular forms "a," "an," and "the" include the plural referents unless the context clearly indicates otherwise.

[0019] 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.

[0020] The terms "CD74" and "CD74 antigen" are used interchangeably herein. Unless specified otherwise, the terms include any variants, isoforms and species homologs of human CD74 that are naturally expressed by cells, or that are expressed by cells transfected with a CD74 gene.

[0021] At least four human isoforms of CD74 are known to exist, including p43, p41, p35 and p33. Borghese *et al*., *Expert Opin. Ther. Targets*, 2011, 15:237-251, incorporated by

reference in its entirety. These isoforms result from alternative transcript splicing and two translation start sites.

[0022] p43 (also known as CD74 isoform 1, isoform a, or "long"; see UniProt entry P04233-1 and NCBI Reference Sequence NP 001020330, each incorporated by reference in its entirety) contains 296 amino acids, with residues 73-296 forming the extracellular portion. Protein constructs of CD74 having the extracellular part of isoform 1 are herein referred to as "variant 1" or "CD74v1."

[0023] p35 (also known as CD74 isoform 2, isoform b or "short"; see UniProt entry P04233-2 and NCBI Reference Sequence NP 004346, each incorporated by reference in its entirety) lacks residues 209-272 from the extracellular domain due to alternative splicing. Protein constructs of CD74 having the extracellular part of isoform 2 are herein referred to as "variant 2" or "CD74v2."

[0024] p41 and p33 arise from an alternative translation start site (48 nucleotides / 16 amino acids downstream) leading to variants lacking the endoplasmic reticulum (ER) retention signal that is present within the eliminated 16 amino acids, but having extracellular domain that is identical to p43 and p35, respectively.

[0025] The sequence of another isoform (known as isoform 3 and isoform c), in which residues 148-160 are replaced and residues 161-296 are lacking, is provided in NP 001020329.

[0026] The sequences of cynomolgus CD74 homologs are provided in, e.g., NCBI Reference Sequence: XP—001099491.2 and NCBI Reference Sequence: XP—002804624.1.

[0027] The term "immunoglobulin" refers to a class of structurally related proteins generally consisting of 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, 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.

[0028] The term "antibody" describes a type of immunoglobulin molecule and is used herein in its broadest sense. An antibody specifically includes monoclonal antibody conjugates, polyclonal antibody conjugates, intact antibody conjugates, and antibody

fragments. Antibody conjugates 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 "CD74 antibody," "anti-CD74 antibody," "CD74 Ab," "CD74-specific antibody" or "anti-CD74 Ab" is an antibody, as described herein, which binds specifically to the antigen CD74.

[0029] 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 confer antigen specificity and binding affinity to 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.

[0030] 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.

[0031] 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.

[0032] 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.

[0033] 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. FIG. 1 provides a comparison of the Kabat and Chothia numbering schemes for CDR-H1. *See* Martin (2010), supra.

[0034] Unless otherwise specified, the numbering scheme used for identification of a particular CDR herein is the Kabat/Chothia numbering scheme. Where these two numbering schemes diverge, the numbering scheme is specified as either Kabat or Chothia.

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.

[0035] The "EU numbering scheme" or "EU index" is generally used when referring to a residue in an antibody heavy chain constant region (e.g., the EU index 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.

[0036] 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.

[0037] "Fv" fragments comprise a non-covalently-linked dimer of one heavy chain variable domain and one light chain variable domain.

[0038] "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 papain digestion of a full-length antibody.

[0039] "$F(ab')_2$" fragments contain two Fab′ fragments joined, near the hinge region, by disulfide bonds. $F(ab')_2$ fragments may be generated, for example, by pepsin digestion of an intact antibody. The F(ab′) fragments can be dissociated, for example, by treatment with ß-mercaptoethanol.

[0040] "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). 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. "scFv-Fc" fragments comprise an scFv attached to an Fc domain. For example, an Fc domain may be attached to the C-terminal 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 is an IgG1 Fc domain (e.g., SEQ ID NO: 289).

[0041] 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.

[0042] 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.

[0043] "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.

[0044] 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.

[0045] 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.

[0046] 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 provided as a solution comprising at least 85%, 90%, 95%, 98%, 99% to 100% by weight of an antibody, the remainder of the weight comprising the weight of other solutes dissolved in the solvent.

[0047] "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 generally 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.

[0048] 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 is similar to the target, such as an excess of non-labeled target. In that case, specific binding is indicated if the binding of the labeled target to a probe is competitively inhibited by the excess non-labeled target.

[0049] 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.

[0050] 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.

[0051] The term "K_D" (M), as used herein, refers to the dissociation equilibrium constant of a particular antibody-antigen interaction. $K_D = k_d/k_a$.

[0052] The term "K_A" (M^{-1}), as used herein, refers to the association equilibrium constant of a particular antibody-antigen interaction. $K_A = k_a/k_d$.

[0053] 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.

[0054] 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., CD74). In one exemplary assay, CD74 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. 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%.

[0055] 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 CD74 variants with different point-mutations.

[0056] 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), or CLUSTALW 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.

[0057] A "conservative substitution" or a "conservative amino acid substitution," refers to the substitution of one or more amino acids with one or more chemically or functionally similar amino acids. 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 following groups of amino acids are considered conservative substitutions for one another.

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

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

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

[0058] 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."

[0059] The term "amino acid" refers to 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).

[0060] 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.

[0061] 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.

[0062] 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.

[0063] 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.

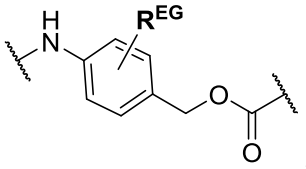
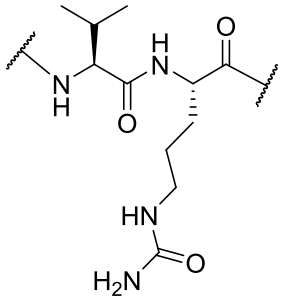
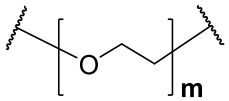
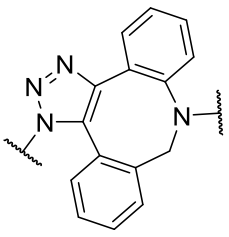
[0064] The term "linker" refers to a molecular moiety that is capable of forming at least two covalent bonds. Typically, a linker is capable of forming at least one covalent bond to an antibody and at least another covalent bond to a payload. In certain embodiments, a linker can form more than one covalent 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.

[0065] "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 the onset of the disease or disorder.

[0066] 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 prevent or ameliorate a disease or the progression of the disease, or result in amelioration of symptoms.

[0067] 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 CD74 antibody, as compared to the growth of the same cells not in contact with a CD74 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.

[0068] 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, goats and sheep. In certain embodiments, the subject is a human. In some embodiments, the subject has cancer, an inflammatory disease or condition, or an autoimmune disease or condition, that can be treated with an antibody provided herein. In some embodiments, the subject is a human that has or is suspected to have cancer, an inflammatory disease or condition, or an autoimmune disease or condition.

[0069] 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.

[0070] 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

[0071] Provided herein are conjugates of antibodies to CD74. The conjugates comprise an antibody to CD74 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.

[0072] 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.

[0073] 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.

[0074] In the conjugates provided herein, the antibody can be any antibody with binding specificity for CD74. The CD74 can be from any species. In certain embodiments, the CD74 is a vertebrate CD74. In certain embodiments, the CD74 is a mammalian CD74. In certain embodiments, the CD74 is human CD74. In certain embodiments, the CD74 is mouse CD74. In certain embodiments, the CD74 is cynomolgus CD74.

[0075] In certain embodiments, the antibody to CD74 competes with an antibody described herein for binding. In certain embodiments, the antibody to CD74 binds to the same epitope as an antibody described herein.

[0076] 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.

[0077] 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, sFv, etc.

[0078] 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). In certain embodiments, the antibody to CD74 is milatuzumab.

[0079] 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.

[0080] 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.

[0081] 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.

[0082] 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.

[0083] 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.

[0084] 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.

[0085] 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.

[0086] 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.

[0087] 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.

[0088] 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.

[0089] 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.

[0090] 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.

[0091] In certain embodiments, the antibodies provided herein comprise one or more non-natural amino acids at one or more positions 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.

[0092] 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-CD74 antibody;

PAY is a payload moiety;

$\mathbf{W^1}$, $\mathbf{W^2}$, $\mathbf{W^3}$, $\mathbf{W^4}$, and $\mathbf{W^5}$ 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.

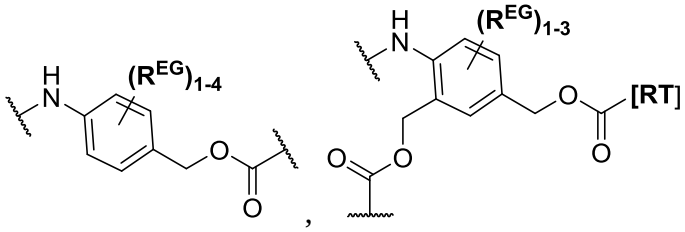
Attaching Groups

[0093] 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 $\mathbf{W^1}$, $\mathbf{W^2}$, $\mathbf{W^3}$, $\mathbf{W^4}$, or $\mathbf{W^5}$. 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.

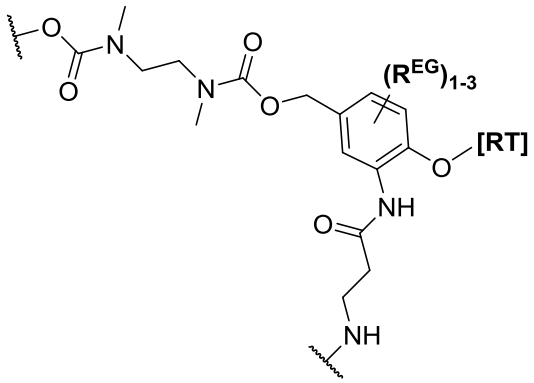
Eliminator Groups

[0094] 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.

[0095] In certain embodiments, the eliminator group is designated **EG** herein. Useful eliminator groups include those described herein. In certain embodiments, the eliminator group is:

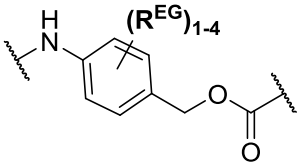


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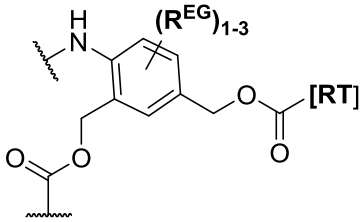


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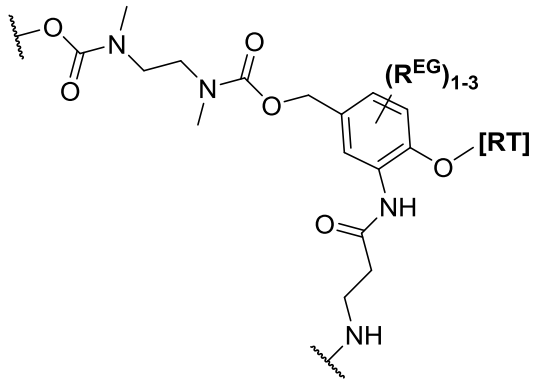
wherein R^{EG} is selected from the group consisting of hydrogen, alkyl, biphenyl, -CF_3, -NO_2, -CN, fluoro, bromo, chloro, alkoxyl, alkylamino, dialkylamino, alkyl-C(O)O-, alkylamino-C(O)- and dialkylaminoC(O)-. In each structure, the phenyl ring can be bound to one, two, three, or in some cases, four 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



. In certain embodiments, the eliminator group is



. In certain embodiments, the eliminator group is

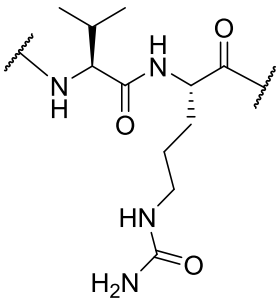


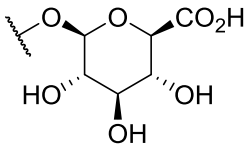
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Release Trigger Groups

[0096] 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.

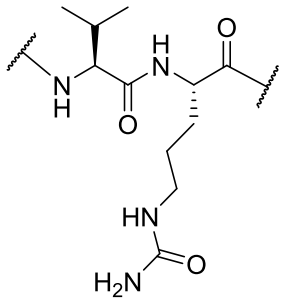
[0097] In certain embodiments, the release trigger group is designated **RT** herein. In certain embodiments, **RT** is divalent and bonded within the backbone of formula (C1). In other embodiments, **RT** is monovalent and bonded to **EG** as depicted above. Useful release trigger groups include those 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:

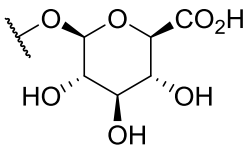


or 

.

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 

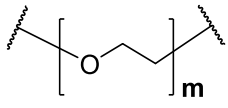
. In certain embodiments, the release trigger group is 

.

Hydrophilic Groups

[0098] 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.

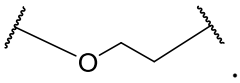
[0099] In certain embodiments, the hydrophilic group is designated **HP** herein. Useful hydrophilic groups include those described herein. In certain embodiments, the hydrophilic group is a divalent poly(ethylene glycol). In certain embodiments, the hydrophilic group is a divalent poly(ethylene glycol) according to the formula:



;

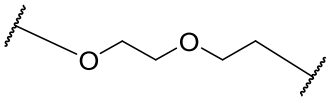
wherein **m** is an integer from 1 to 12, optionally 1 to 4, optionally 2 to 4.

[00100] In some embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



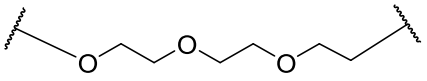
.

[00101] In some other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



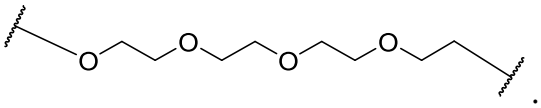
.

[00102] In other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:



.

[00103] In other embodiments, the hydrophilic group is a divalent poly(ethylene glycol) having the following formula:

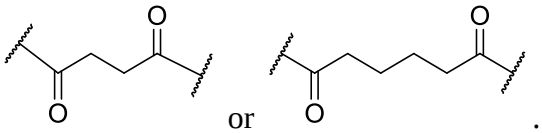


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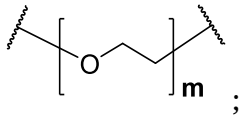
Spacer Groups

[00104] 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. The spacer group can also stabilize the conjugating group.

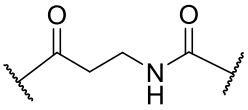
[00105] In certain embodiments, the spacer group is designated **SP** herein. Useful spacer groups include those described herein. In certain embodiments, the spacer group is:



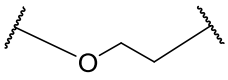
In certain embodiments, the spacer group, $\mathbf{W^4}$, and the hydrophilic group combine to form a divalent poly(ethylene glycol) according to the formula:



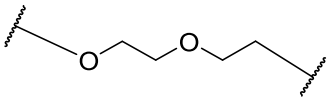
wherein **m** is an integer from 1 to 12, optionally 1 to 4, optionally 2 to 4.

[00106] In some embodiments, the **SP** is .

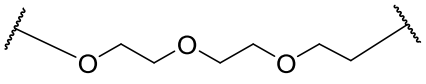
[00107] In some embodiments, the divalent poly(ethylene glycol) has the following formula:

.

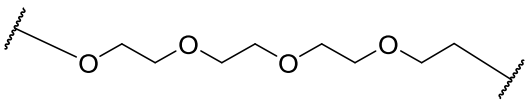
[00108] In some other embodiments, the divalent poly(ethylene glycol) has the following formula:

.

[00109] In other embodiments, the divalent poly(ethylene glycol) has the following formula:

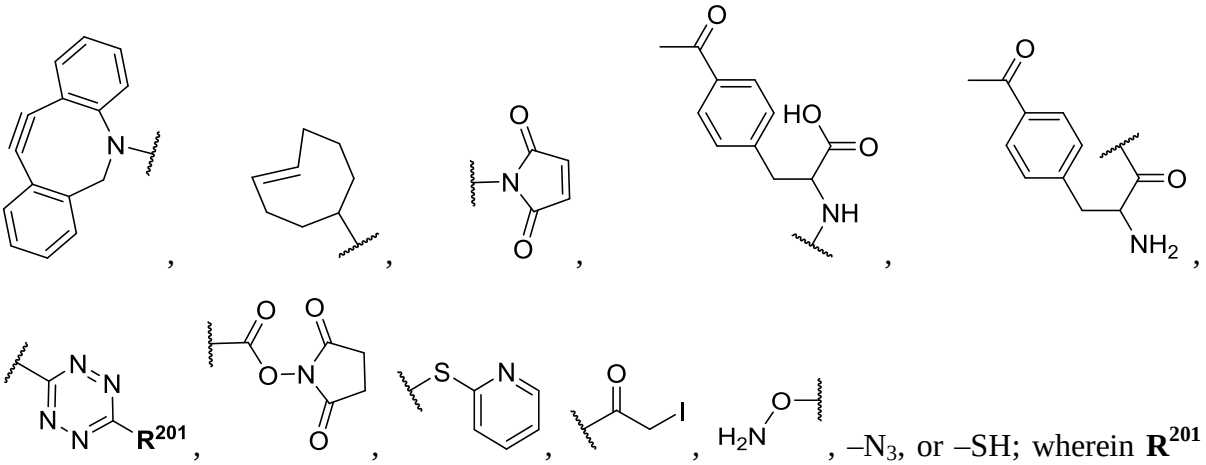
.

[00110] In other embodiments, the divalent poly(ethylene glycol) has the following formula:



Conjugating Groups and Residues Thereof

[00111] Conjugating groups facilitate conjugation of the compounds described herein to a second compound, such as a targeting moiety. 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:

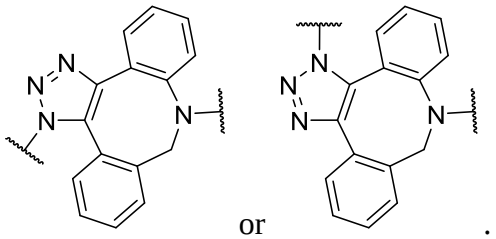


, –N_3, or –SH; wherein $\mathbf{R^{201}}$ is lower alkyl. In an embodiment, $\mathbf{R^{201}}$ is methyl, ethyl, or propyl. In an embodiment, $\mathbf{R^{201}}$ is methyl. Additional conjugating groups are described in, for example, U.S. Patent Publication No. 2014/0356385, U.S. Patent Publication No. 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.

[00112] 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.

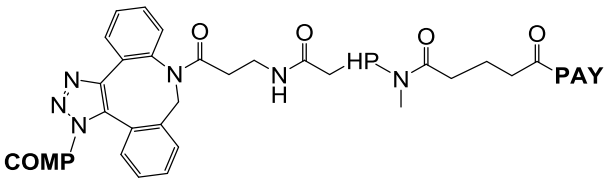
[00113] In certain embodiments when a conjugate is formed through a [3+2] alkyne-azide cycloaddition reaction, the divalent residue of the conjugating group comprises a triazole ring or fused cyclic group comprising a triazole ring. In certain embodiment when a

conjugate is formed through a [3+2] alkyne-azide cycloaddition reaction, the divalent residue of the conjugating group is:

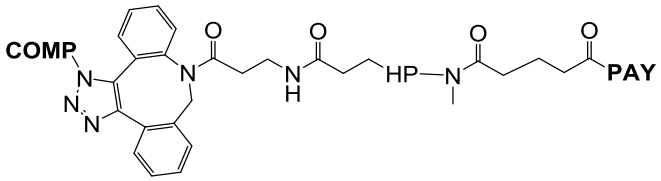


or .

[00114] In an embodiment, provided herein is a conjugate according to any of the following formulas, where **COMP** indicates a residue of the anti-CD74 anibody and **PAY** indicates a payload moiety:

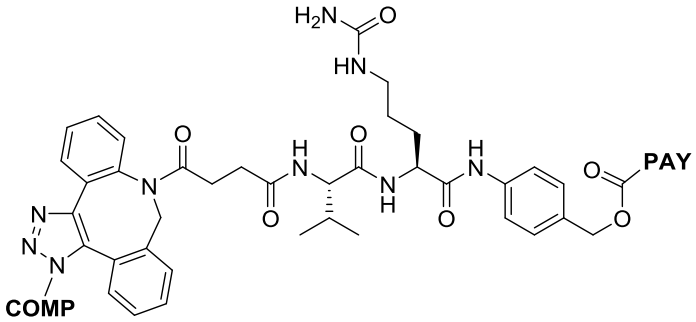


or

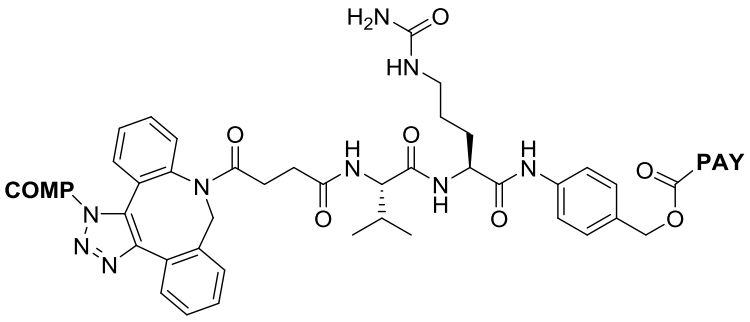


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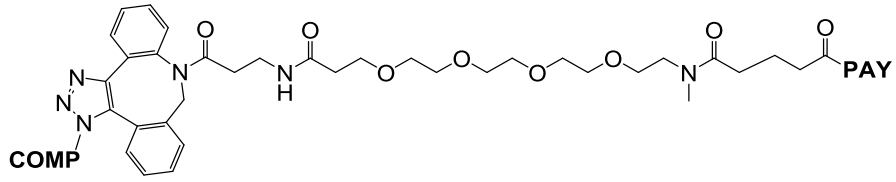
[00115] In an embodiment, provided herein is a conjugate according to any of Formulas 101a-104b, where **COMP** indicates a residue of the anti-CD74 anibody and **PAY** indicates a payload moiety:



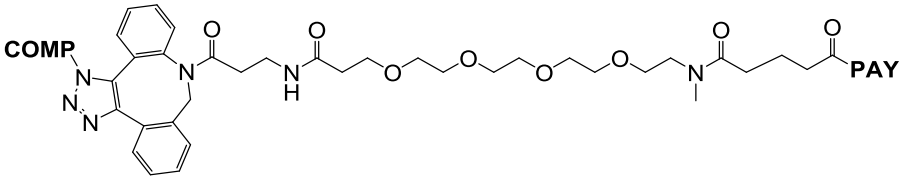
(101a)



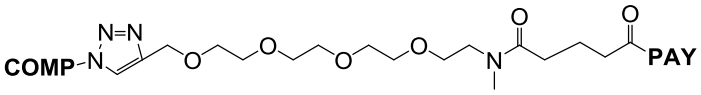
(101b)



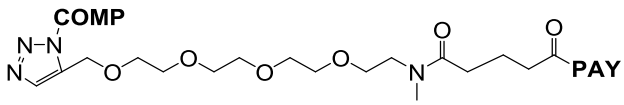
(102a)



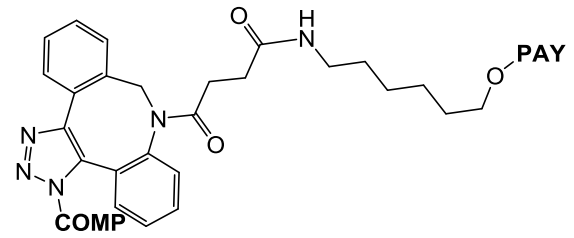
(102b)



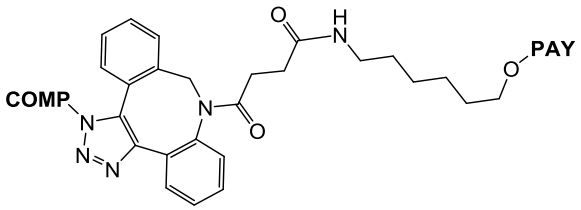
(103a)



(103b)



(104a)



(104b).

[00116] In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (30), below. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (30), below, at heavy chain position 404 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (30), below, at heavy chain position 241 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (30), below, at heavy chain position 222 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (30), below, at light chain position 7 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.

[00117] In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (56), below. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (56), below, at heavy chain position 404 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (56), below, at heavy chain position 241 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (56), below, at heavy chain position 222 according to the EU numbering system. In particular embodiments, provided herein are anti-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates a non-natural amino acid residue according to Formula (56), below, at light chain position 7 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.

[00118] In particular embodiments, provided herein are anti-CD74 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-CD74 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-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates the 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-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates the 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-CD74 conjugates according to any of Formulas 101a-104b wherein **COMP** indicates the non-

natural amino acid residue para-azido-L-phenylalanine at light chain position 7 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.

3. Payloads

[00119] 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.

[00120] 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.

[00121] 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.

[00122] 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.

[00123] 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.

[00124] 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.

[00125] 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).

[00126] 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.

[00127] In some embodiments, the payload is not a radioisotope. In some embodiments, the payload is not radioactive.

[00128] 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.

[00129] 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, 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.

[00130] 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).

[00131] 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.

[00132] Suitable cyclooxygenase inhibitors include meclofenamic acid, mefenamic acid, carprofen, diclofenac, diflunisal, fenbufen, fenoprofen, indomethacin, ketoprofen, nabumetone, sulindac, tenoxicam and tolmetin.

[00133] 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.

[00134] 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.

[00135] 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.

[00136] 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.

[00137] 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 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, 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.

[00138] 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); and (x) pharmaceutically acceptable salts, acids and derivatives of any of the above. Other anti-angiogenic agents include 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).

[00139] 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.

[00140] 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.

[00141] 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.

[00142] 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.

[00143] 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.

[00144] 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.

[00145] 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.

[00146] 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.

[00147] 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. 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.

[00148] 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.

[00149] 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

[00150] 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.

[00151] 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.

[00152] 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.

[00153] 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.

[00154] 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.

[00155] 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.

[00156] 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.

[00157] 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.

[00158] 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.

[00159] 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).

[00160] 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) and glycine-glycine-glycine (gly-gly-gly).

[00161] 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).

[00162] 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 val-cit-PAB linkers.

[00163] 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.

5. Antibody Specificity

[00164] The conjugates comprise antibodies that selectively bind human CD74. In some aspects, the antibody selectively binds to human CD74 isoform 1. In some aspects, the antibody selectively binds to human CD74 isoform 2. In some aspects, the antibody may selectively bind to more than one CD74 isoform, for example, both human CD74 isoforms 1 and 2. In some aspects, the antibody may selectively bind to one or more CD74 isoforms with the same extracellular domain as isoforms 1 and 2, such as p41 and p33, respectively.

[00165] In some embodiments, the antibody binds to homologs of human CD74. In some aspects, the antibody binds to a homolog of human CD74 from a species selected from monkeys, dogs, cats, mice, rats, cows, horses, goats or sheep. In some aspects, the homolog is a cynomolgus monkey homolog.

[00166] In some embodiments, the antibodies have higher melting temperatures than other anti-CD74 antibodies. In some aspects, the Tm2 of the antibody is higher than other anti-CD74 antibodies. The Tm2 represents the melting temperature of the Fab domain of an IgG. A higher Tm2 therefore promotes stability of the antibody binding site. Such improved stability can lead to better stability of the antibody during storage, as well as improved yield during manufacturing.

[00167] 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.

[00168] In some embodiments, the antibody has one or more CDRs having particular lengths, in terms of the number of amino acid residues. In some aspects, the Chothia CDR-H1 of the antibody is seven residues in length. In some embodiments, the Kabat CDR-H1 of the antibody is five residues in length. In some aspects, the Chothia CDR-H2 of the antibody is six residues in length. In some embodiments, the Kabat CDR-H2 of the antibody is seventeen residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is eleven residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is twelve residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is thirteen residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is fourteen residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is fifteen residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is more than fifteen residues in length. In some aspects, the Chothia/Kabat CDR-H3 of the antibody is up to twenty-five residues in length.

[00169] In some aspects, the Kabat/Chothia CDR-L1 of the antibody is eleven residues in length. In some aspects, the Kabat/Chothia CDR-L1 of the antibody is twelve residues in length. In some aspects, the Kabat/Chothia CDR-L2 of the antibody is seven residues in length. In some aspects, the Kabat/Chothia CDR-L3 of the antibody is nine residues in length.

[00170] In some embodiments, the antibody comprises a light chain. In some aspects, the light chain is selected from a kappa light chain and a lambda light chain.

[00171] In some embodiments, the antibody comprises a heavy chain. In some aspects, the heavy chain is selected from IgA, IgD, IgE, IgG, and IgM. In some aspects, the heavy chain is selected from IgG1, IgG2, IgG3, IgG4, IgA1, and IgA2.

[00172] In some embodiments, the antibody is an antibody fragment. In some aspects, 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.

[00173] In some embodiments, the antibody is a monoclonal antibody.

[00174] In some embodiments, the antibody is a chimeric, humanized, or human antibody.

[00175] 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.

[00176] In some embodiments, the antibody is internalized by a cell after binding.

[00177] In some embodiments, the antibody inhibits the binding of CD74 to its ligands. In some aspects, the antibody inhibits the binding of CD74 to macrophage migration inhibitory factor (MIF).

[00178] In some embodiments, the antibody is one that is described in WO2016/014434 A2, which is incorporated herein by reference in its entirety.

5.1. CDR-H3 Sequences

[00179] In some embodiments, the antibody comprises a CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 129-160.

[00180] In some aspects, the CDR-H3 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 157-160. In some aspects, the CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 157. In some aspects, the CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 158. In some aspects, the CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 159. In some aspects, the CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 160.

5.2. V_H Sequences Comprising Illustrative CDRs

[00181] 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.

5.2.1. V_H Sequences Comprising Illustrative Kabat CDRs

[00182] 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

[00183] 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: 129-156.

5.2.1.2.Kabat CDR-H2

[00184] 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: 97-124.

5.2.1.3.Kabat CDR-H1

[00185] 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: 33-60.

5.2.1.4.Kabat CDR-H3 + Kabat CDR-H2

[00186] 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: 129-156 and a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 97-124. 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: 230-251 and 273-280.

5.2.1.5.Kabat CDR-H3 + Kabat CDR-H1

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: 129-156 and a Kabat CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 33-60. 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: 230-251 and 273-280.

5.2.1.6.Kabat CDR-H1 + Kabat CDR-H2

[00187] 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: 33-60 and a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 97-124. 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: 230-251 and 273-280.

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

[00188] 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: 33-60, a Kabat CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 97-124, and a Kabat CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 129-156. 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: 230-251 and 273-280.

5.2.1.8. Excluded V_H Sequences Comprising Kabat CDRs

[00189] In some embodiments, the V_H sequences provided herein do not comprise certain Kabat CDR-H3, CDR-H2, and/or CDR-H1 sequences.

[00190] In some aspects, the Kabat CDR-H3 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 157-160. In some aspects, the Kabat CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 157. In some aspects, the Kabat CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 158. In some aspects, the Kabat CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 159. In some aspects, the Kabat CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 160.

[00191] In some aspects, the Kabat CDR-H2 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 125-128. In some aspects, the Kabat CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 125. In some aspects, the Kabat CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 126. In some aspects, the Kabat CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 127. In some aspects, the Kabat CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 128.

[00192] In some aspects, the Kabat CDR-H1 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 61-64. In some aspects, the Kabat CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID

NO: 61. In some aspects, the Kabat CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 62. In some aspects, the Kabat CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 63. In some aspects, the Kabat CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 64.

5.2.2. V_H Sequences Comprising Illustrative Chothia CDRs

[00193] 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

[00194] 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: 129-156.

5.2.2.2.Chothia CDR-H2

[00195] 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: 65-92.

5.2.2.3.Chothia CDR-H1

[00196] 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: 1-28.

5.2.2.4.Chothia CDR-H3 + Chothia CDR-H2

[00197] 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: 129-156 and a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 65-92. 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: 230-251 and 273-280.

5.2.2.5.Chothia CDR-H3 + Chothia CDR-H1

[00198] 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: 129-156 and a Chothia CDR-H1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 1-28. 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: 230-251 and 273-280.

5.2.2.6.Chothia CDR-H1 + Chothia CDR-H2

[00199] 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: 1-28 and a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 65-92. 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: 230-251 and 273-280.

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

[00200] 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: 1-28, a Chothia CDR-H2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 65-92, and a Chothia CDR-H3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 129-156. 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: 230-251 and 273-280.

5.2.2.8. Excluded V_H Sequences Comprising Chothia CDRs

[00201] In some embodiments, the V_H sequences provided herein do not comprise certain Chothia CDR-H3, CDR-H2, and/or CDR-H1 sequences.

[00202] In some aspects, the Chothia CDR-H3 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 157-160. In some aspects, the Chothia CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 157. In some aspects, the Chothia CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 158. In some aspects, the Chothia CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 159. In some aspects, the Chothia CDR-H3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 160.

[00203] In some aspects, the Chothia CDR-H2 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 93-96. In some aspects, the Chothia CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 93. In some aspects, the Chothia CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 94. In some aspects, the Chothia CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 95. In some aspects, the Chothia CDR-H2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 96.

[00204] In some aspects, the Chothia CDR-H1 sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 29-32. In some aspects, the Chothia CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 29. In some aspects, the Chothia CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 30. In some aspects, the Chothia CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 31. In some aspects, the Chothia CDR-H1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 32.

5.3. V_H Sequences

[00205] In some embodiments, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 230-251 and 273-280. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 230. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 231. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 232. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 233. In some aspects, the

antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 234. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 235. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 236. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 237. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 238. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 239. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 240. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 241. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 242. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 243. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 244. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 245. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 246. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 247. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 248. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 249. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 250. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 251. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 273. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 274. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 275. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 276. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 277. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 278. In some aspects, the antibody comprises a V_H sequence

comprising, consisting of, or consisting essentially of SEQ ID NO: 279. In some aspects, the antibody comprises a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 280.

5.3.1. Excluded V_H Sequences

[00206] In some embodiments, the V_H sequences provided herein do not comprise certain V_H sequences.

[00207] In some aspects, the V_H sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 252-255. In some aspects, the V_H sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 252. In some aspects, the V_H sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 253. In some aspects, the V_H sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 254. In some aspects, the V_H sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 255.

5.4. CDR-L3 Sequences

[00208] In some embodiments, the antibody comprises a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 201-219.

[00209] In some aspects, the CDR-L3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 220.

5.5. V_L Sequences Comprising Illustrative CDRs

[00210] 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

[00211] 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: 201-219.

5.5.2. CDR-L2

[00212] 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: 181-199.

5.5.3. CDR-L1

[00213] 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: 161-179.

5.5.4. CDR-L3 + CDR-L2

[00214] 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: 201-219 and a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-199. 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: 256-270 and 281-288.

5.5.5. CDR-L3 + CDR-L1

[00215] 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: 201-219 and a CDR-L1 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 161-179. 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: 256-270 and 281-288.

5.5.6. CDR-L1 + CDR-L2

[00216] 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: 161-179 and a CDR-L2 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 181-199. 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: 256-270 and 281-288.

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

[00217] 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: 161-179, a CDR-L2 sequence comprising, consisting of, or consisting

essentially of a sequence selected from SEQ ID NOs: 181-199, and a CDR-L3 sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 201-219. 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: 256-270 and 281-288.

5.5.8. Excluded V_L Sequences Comprising CDR-Ls

[00218] In some embodiments, the V_L sequences provided herein do not comprise certain CDR-L3, CDR-L2, and/or CDR-L1 sequences.

[00219] In some aspects, the CDR-L3 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 220.

[00220] In some aspects, the CDR-L2 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 200.

[00221] In some aspects, the CDR-L1 sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 180.

5.6. V_L Sequences

[00222] In some embodiments, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of a sequence selected from SEQ ID NOs: 256-270 and 281-288. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 256. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 257. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 258. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 259. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 260. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 261. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 262. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 263. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 264. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 265. In some aspects, the antibody comprises a V_L sequence comprising, consisting

of, or consisting essentially of SEQ ID NO: 266. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 267. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 268. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 269. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 270. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 281. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 282. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 283. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 284. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 285. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 286. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 287. In some aspects, the antibody comprises a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NO: 288.

5.6.1. Excluded V_L Sequences

[00223] In some embodiments, the V_L sequences provided herein do not comprise certain V_L sequences.

[00224] In some aspects, the V_L sequence does not comprise, consist of, or consist essentially of a sequence selected from SEQ ID NOs: 271-272. In some aspects, the V_L sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 271. In some aspects, the V_L sequence does not comprise, consist of, or consist essentially of SEQ ID NO: 272.

5.7. Pairs

5.7.1. CDR-H3 – CDR-L3 Pairs

[00225] 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.

[00226] In some aspects, the CDR-H3 sequence is a CDR-H3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 129-156 and the CDR-L3 sequence is

a CDR-L3 sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 201-219.

5.7.1.1. Excluded CDR-H3 – CDR-L3 Pairs

[00227] In some embodiments, the CDR-H3 – CDR-L3 pairs provided herein do not comprise certain CDR-H3 – CDR-L3 pairs.

[00228] In some aspects, the CDR-H3 sequence is not SEQ ID NO: 157, and the CDR-L3 sequence not SEQ ID NO: 220. In some aspects, the CDR-H3 sequence is not SEQ ID NO: 158, and the CDR-L3 sequence not SEQ ID NO: 220. In some aspects, the CDR-H3 sequence is not SEQ ID NO: 159, and the CDR-L3 sequence not SEQ ID NO: 220. In some aspects, the CDR-H3 sequence is not SEQ ID NO: 160, and the CDR-L3 sequence not SEQ ID NO: 220.

5.7.2. $V_H – V_L$ Pairs

[00229] In some embodiments, the antibody comprises a V_H sequence and a V_L sequence.

[00230] In some aspects, the V_H sequence is a V_H sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 230-251 and 273-280, and the V_L sequence is a V_L sequence comprising, consisting of, or consisting essentially of SEQ ID NOs: 256-270 and 281-288.

[00231] In some aspects, the V_H sequence is SEQ ID NO: 230, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00232] In some aspects, the V_H sequence is SEQ ID NO: 231, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00233] In some aspects, the V_H sequence is SEQ ID NO: 232, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00234] In some aspects, the V_H sequence is SEQ ID NO: 233, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00235] In some aspects, the V_H sequence is SEQ ID NO: 234, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00236] In some aspects, the V_H sequence is SEQ ID NO: 235, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00237] In some aspects, the V_H sequence is SEQ ID NO: 236, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00238] In some aspects, the V_H sequence is SEQ ID NO: 237, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00239] In some aspects, the V_H sequence is SEQ ID NO: 238, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00240] In some aspects, the V_H sequence is SEQ ID NO: 239, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00241] In some aspects, the V_H sequence is SEQ ID NO: 240, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00242] In some aspects, the V_H sequence is SEQ ID NO: 241, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00243] In some aspects, the V_H sequence is SEQ ID NO: 242, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00244] In some aspects, the V_H sequence is SEQ ID NO: 243, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00245] In some aspects, the V_H sequence is SEQ ID NO: 244, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00246] In some aspects, the V_H sequence is SEQ ID NO: 245, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00247] In some aspects, the V_H sequence is SEQ ID NO: 246, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00248] In some aspects, the V_H sequence is SEQ ID NO: 247, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00249] In some aspects, the V_H sequence is SEQ ID NO: 248, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00250] In some aspects, the V_H sequence is SEQ ID NO: 249, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00251] In some aspects, the V_H sequence is SEQ ID NO: 250, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00252] In some aspects, the V_H sequence is SEQ ID NO: 251, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00253] In some aspects, the V_H sequence is SEQ ID NO: 273, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00254] In some aspects, the V_H sequence is SEQ ID NO: 274, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00255] In some aspects, the V_H sequence is SEQ ID NO: 275, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00256] In some aspects, the V_H sequence is SEQ ID NO: 276, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00257] In some aspects, the V_H sequence is SEQ ID NO: 277, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00258] In some aspects, the V_H sequence is SEQ ID NO: 278, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00259] In some aspects, the V_H sequence is SEQ ID NO: 279, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

[00260] In some aspects, the V_H sequence is SEQ ID NO: 280, and the V_L sequence is selected from SEQ ID NOs: 256-270 and 281-288.

5.7.2.1. Excluded $V_H – V_L$ Pairs

[00261] In some embodiments, the $V_H – V_L$ pairs provided herein do not comprise certain $V_H – V_L$ pairs.

[00262] In some aspects, the V_H sequence is not selected from SEQ ID NOs: 252-255, and the V_L sequence is not selected from SEQ ID NOs: 271-272.

[00263] In some aspects, the V_H sequence is not SEQ ID NO: 252, and the V_L sequence is not selected from SEQ ID NO: 271-272. In some aspects, the V_L sequence is not SEQ ID NO: 271. In some aspects, the V_L sequence is not SEQ ID NO: 272.

[00264] In some aspects, the V_H sequence is not SEQ ID NO: 253, and the V_L sequence is not selected from SEQ ID NO: 271-272. In some aspects, the V_L sequence is not SEQ ID NO: 271. In some aspects, the V_L sequence is not SEQ ID NO: 272.

[00265] In some aspects, the V_H sequence is not SEQ ID NO: 254, and the V_L sequence is not selected from SEQ ID NO: 271-272. In some aspects, the V_L sequence is not SEQ ID NO: 271. In some aspects, the V_L sequence is not SEQ ID NO: 272.

[00266] In some aspects, the V_H sequence is not SEQ ID NO: 255, and the V_L sequence is not selected from SEQ ID NO: 271-272. In some aspects, the V_L sequence is not SEQ ID NO: 271. In some aspects, the V_L sequence is not SEQ ID NO: 272.

6. Thermostability

[00267] In some embodiments, the antibody is characterized by particular thermostability parameters. The thermostability of an antibody may be characterized by measuring its melting temperatures. The melting temperatures include Tm1 and Tm2. Tm1

represents the melting of the Fc domain of an IgG, while Tm2 represents the melting of the Fab domain of an IgG.

[00268] In some embodiments, the Tm2 of the antibody is at least 75°C, 75.5°C, 76°C, 76.5°C, 77°C, 77.5°C, 78°C, 78.5°C, or 79°C. In some embodiments, the Tm2 of the antibody is between about 75°C and about 80°C. In some embodiments, the Tm2 of the antibody is between about 76°C and about 79°C. In some embodiments, the Tm2 of the antibody is between about 77°C and about 78°C. In some aspects, the Tm2s described above are for aglycosylated versions of the antibody.

[00269] In some embodiments, the Tm1 of the antibody is between about 59°C and about 62.2°C. In some embodiments, the Tm1 of the antibody is less than 62.2°C. In some embodiments, the Tm1 of the antibody is less than 61°C. In some embodiments, the Tm1 of the antibody is less than 60°C. In some aspects, the Tm1s described above are for aglycosylated versions of the antibody.

7. Affinity

[00270] In some embodiments, the affinity of the antibody for CD74, 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 conjugate is between about 10^{-9} M and 10^{-11} M. In some embodiments, the affinity of the antibody conjugate is between about 10^{-10} M and 10^{-11} M. In some embodiments, the affinity of the antibody is between about 1.08×10^{-7} M and 9.57×10^{-10} M. In some embodiments, the affinity of the antibody is 2.52×10^{-10} M, or less. In some embodiments, the affinity of the antibody is about 2.52×10^{-10} M. In some embodiments, the affinity of the antibody is about 3.54×10^{-10} M. In some embodiments, the affinity of the antibody is between about 2.52×10^{-10} M and about 3.54×10^{-10} M. In some aspects, the K_D is determined at 25°C.

[00271] 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 between about 10^5 $M^{-1}\times sec^{-1}$ and about 10^6 $M^{-1}\times sec^{-1}$.

In some embodiments the antibody has a k_a of between about 1.66×10^5 $M^{-1}\times sec^{-1}$ and about 1.07×10^6 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of about 3.09×10^5 $M^{-1}\times sec^{-1}$, or more. In some embodiments the antibody has a k_a of about 3.09×10^5 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a of about 3.38×10^5 $M^{-1}\times sec^{-1}$. In some embodiments the antibody has a k_a between about 3.09×10^5 $M^{-1}\times sec^{-1}$ and about 3.38×10^5 $M^{-1}\times sec^{-1}$. In some aspects, the k_a is determined at 25°C.

[00272] 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^{-5} sec^{-1} or less. In some embodiments the antibody has a k_d of between about 10^{-4} sec^{-1} and about 10^{-5} sec^{-1}. In some embodiments the antibody has a k_d of between about 2.35×10^{-4} sec^{-1} and about 7.10×10^{-5} sec^{-1}. In some embodiments the antibody has a k_d of about 7.77×10^{-5} sec^{-1}, or less. In some embodiments the antibody has a k_d of about 7.77×10^{-5} sec^{-1}. In some embodiments the antibody has a k_d of about 1.20×10^{-4} sec^{-1}. In some embodiments the antibody has a k_d between about 1.20×10^{-4} sec^{-1} and about 7.77×10^{-5} sec^{-1}. In some aspects, the k_d is determined at 25°C.

8. Glycosylation Variants

[00273] 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."

[00274] "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.

[00275] "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.

[00276] 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 conjugate.

9. Fc Variants

[00277] 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 conjugate-directed complement-mediated cytotoxicity (ADCC). Numerous substitutions or substitutions or deletions with altered effector function are known in the art.

[00278] 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.

[00279] 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. 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.

[00280] 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.

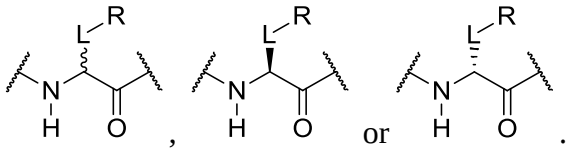
[00281] Complement activation assays include those described, for example, in Gazzano-Santoro *et al., J. Immunol. Methods*, 1996, 202:163-171; Cragg *et al., Blood,* 2003, 101:1045-1052; and Cragg and Glennie, *Blood*, 2004, 103:2738-2743.

[00282] 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.

10. Modified Amino Acids

[00283] 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.

[00284] 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.

[00285] 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.

[00286] 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.

[00287] 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.

[00288] 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.

[00289] 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.

[00290] 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-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.

[00291] 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.

[00292] 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-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.

[00293] In particular embodiments, the non-natural amino acids are selected from p-acetyl- phenylalanine, p-ethynyl-phenylalanine, p-propargyloxyphenylalanine, 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.

[00294] 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.

[00295] 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 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.

[00296] 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

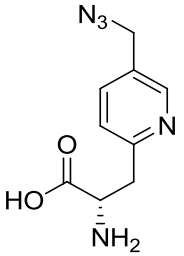
physiological conditions, and further preferably such incorporation can occur via an in vivo system; and (4) the non-natural amino acid includes an oxime functional group or a functional group that can be transformed into an oxime group by reacting with a reagent, preferably under conditions that do 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.

[00297] 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.

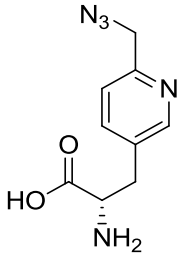
[00298] In some embodiments, non-natural amino acids comprise a saccharide moiety. Examples of such amino acids include N-acetyl-L-glucosaminyl-L-serine, N-acetyl-L-galactosaminyl-L-serine, N-acetyl-L-glucosaminyl-L-threonine, N-acetyl-L-glucosaminyl-L-asparagine and O-mannosaminyl-L-serine. Examples 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.

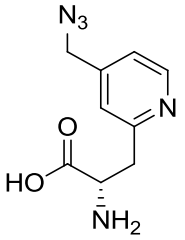
[00299] In particular embodiments, the non-natural amino acid is selected from the group consisting of:



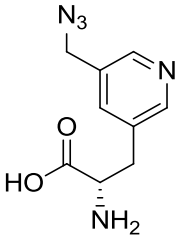
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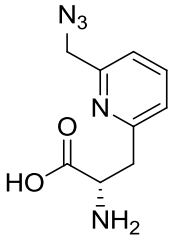
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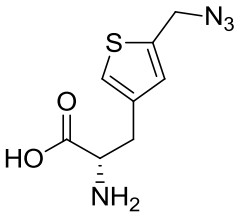
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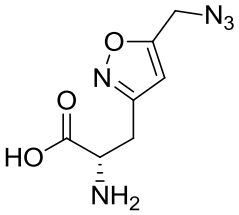
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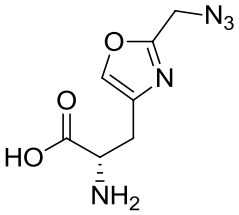
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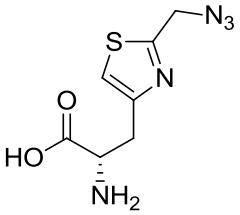
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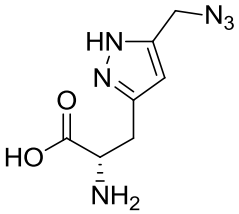
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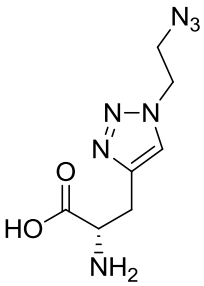
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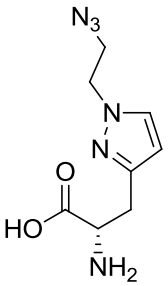
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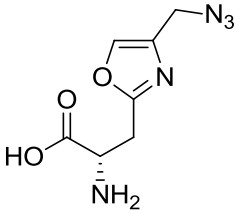
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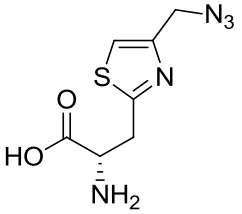
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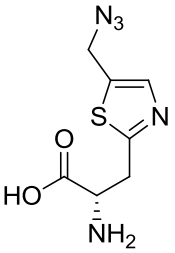
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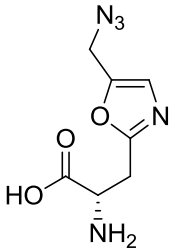
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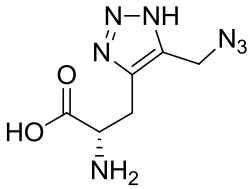
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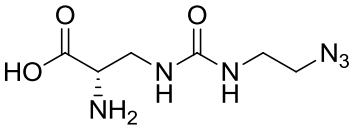
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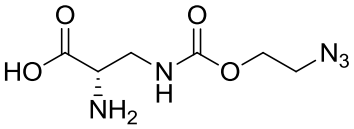
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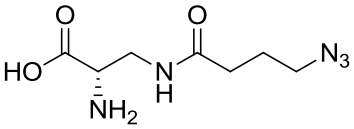
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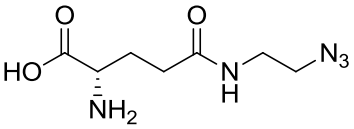
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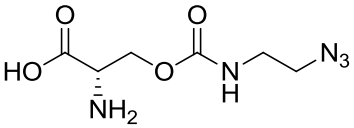
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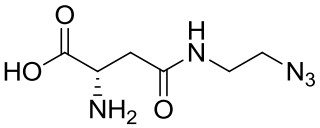
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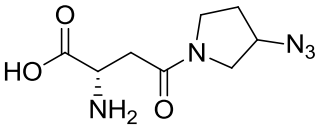
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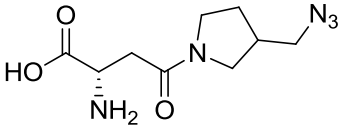
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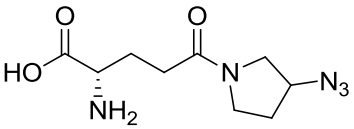
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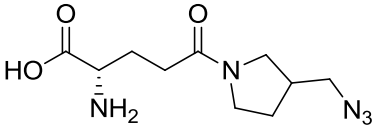
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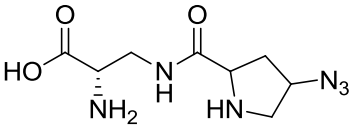
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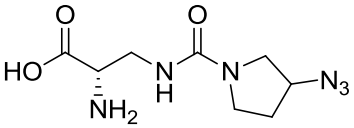
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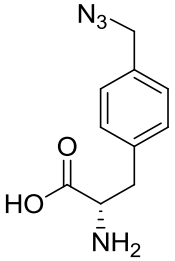
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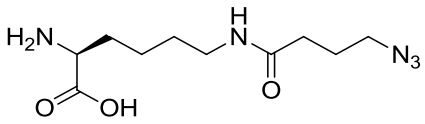
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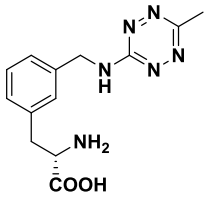
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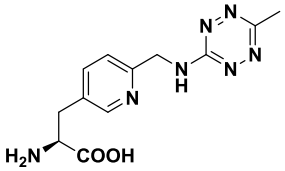
and (40);

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.

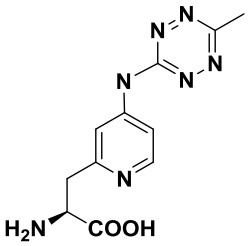
[00300] In certain embodiments, the modified amino acid is according to any of formulas 51-60:



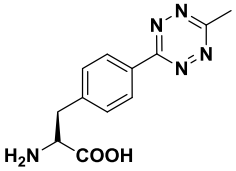
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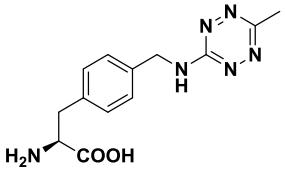
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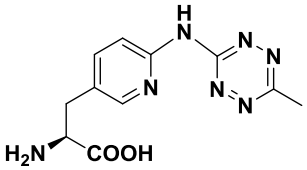
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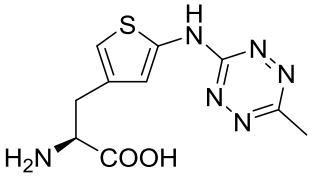
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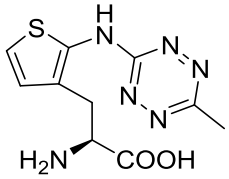
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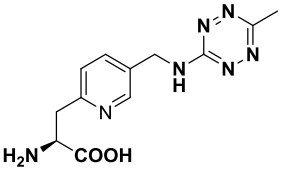
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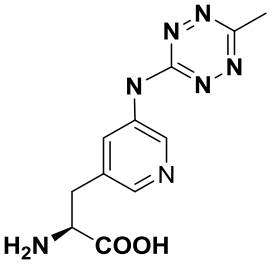
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(60)

or a salt thereof.

[00301] In certain embodiments, the non-natural amino acid is selected from the group consisting of compounds 30, 53, 56, 59, and 60 above. In certain embodiments, the non-natural amino acid is compound 30. In certain embodiments, the non-natural amino acid is compound 56.

11. Preparation of Antibody Conjugates

11.1. Antigen Preparation

[00302] The CD74 antigen to be used for production of antibodies may be intact CD74 or a fragment of CD74. Other forms of CD74 useful for generating antibodies will be apparent to those skilled in the art.

11.2. Monoclonal Antibodies

[00303] Monoclonal antibodies may be obtained, for example, using the hybridoma method first described by Kohler *et al*., *Nature*, 1975, 256:495-497, and/or by recombinant DNA methods (*see e.g.,* U.S. Patent No. 4,816,567). 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.

[00304] 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.

[00305] 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.

[00306] 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.

[00307] 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.

[00308] 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.

11.3. Humanized Antibodies

[00309] Humanized antibodies may be generated by replacing most, or all, of the structural portions of a 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.

11.4. Human Antibodies

[00310] 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. 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). 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). Human antibodies may also be derived from yeast-based libraries (*see e.g.,* U.S. Patent No. 8,691,730).

11.5. Conjugation

[00311] 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 paylaod to the linker. The resulting payload-linker is contacted with an antibody under conditions suitable for forming a bond from the paylaod-linker to the antibody to form an antibody-linker-payload conjugate. Exemplary conditions are described in the examples below.

12. Vectors, Host Cells, and Recombinant Methods

[00312] The invention also provides isolated nucleic acids encoding anti-CD74 antibody conjugates, vectors and host cells comprising the nucleic acids, and recombinant techniques for the production of the antibodies.

[00313] For recombinant production of the antibody, the nucleic acid 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.

[00314] 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.

[00315] Illustrative examples of suitable host cells are provided below. these host cells are not meant to be limiting.

[00316] 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.

[00317] In addition to prokaryotes, eukaryotic microbes such as filamentous fungi or yeast are also suitable cloning or expression hosts for anti-CD74 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 *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*).

[00318] 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.

[00319] The host cells used to produce the anti-CD74 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.

[00320] 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.

[00321] 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.

[00322] 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.

[00323] 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.

[00324] 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.

[00325] 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). Protein G is useful for all mouse isotypes and for human γ3 (Guss *et al*., *EMBO J.*, 1986, 5:1567-1575).

[00326] 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.

[00327] 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.

[00328] 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 2.5-4.5, generally performed at low salt concentrations (e.g., from about 0-0.25 M salt).

13. Pharmaceutical Compositions and Methods of Administration

[00329] 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.

[00330] The methods provided herein encompass administering pharmaceutical compositions comprising at least one antibody 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*.

[00331] In clinical practice the pharmaceutical compositions or antibody conjugates provided herein may be administered by any route known in the art. In certain embodiments, a pharmaceutical composition or antibody provided herein is administered parenterally.

[00332] 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.

[00333] 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 antibody conjugates.

[00334] Typical pharmaceutical compositions and dosage forms comprise one or more excipients. Suitable excipients are well-known to those skilled in the art of pharmacy, and 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.

[00335] 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.

[00336] Further encompassed herein are anhydrous pharmaceutical compositions and dosage forms comprising an antibody, since water can facilitate the degradation of some antibody conjugates.

[00337] 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.

[00338] An anhydrous pharmaceutical composition should 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.

[00339] Further provided are pharmaceutical compositions and dosage forms that comprise one or more excipients that reduce the rate by which an antibody will decompose. Such antibody conjugates, which are referred to herein as "stabilizers," include, but are not limited to, antioxidants such as ascorbic acid, pH buffers, or salt buffers.

13.1. Parenteral Dosage Forms

[00340] 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.

[00341] 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.

[00342] Excipients that increase the solubility of one or more of the antibody conjugates disclosed herein can also be incorporated into the parenteral dosage forms.

13.2. Dosage and Unit Dosage Forms

[00343] 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, stage of the infection and other factors specific to the subject to be treated.

[00344] The amount of the antibody 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.

[00345] 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 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 mg/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.

[00346] 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 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.

[00347] 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 antibody conjugates 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.

[00348] In certain embodiments, treatment or prevention can be initiated with one or more loading doses of an antibody or composition provided herein followed by one or more maintenance doses.

[00349] In certain embodiments, a dose of an antibody 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.

[00350] 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. Therapeutic Applications

[00351] For therapeutic applications, the antibody conjugates of the invention are 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 of the invention 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.

[00352] The antibody conjugates provided herein may be useful for the treatment of any disease or condition involving upregulation of CD74. Upregulation of CD74 expression has been observed in cancers and autoimmune disease (Borghese *et al*., *Exp. Op. Ther. Targets*, 2011, 15:237-251, incorporated by reference in its entirety), as well as in infection (Hofman *et al*., *Modern Pathology*, 2007, 20:974-989, incorporated by reference in its entirety) and inflammatory conditions (Vera *et al*., *Exp. Biol. & Med.*, 2008, 233:620-626, incorporated by reference in its entirety). CD74 is known to be expressed at moderate to high levels in multiple myeloma. Burton *et al*., *Clin. Cancer Res.*, 2004, 10:6606-6611, incorporated by reference in its entirety. CD74 expression is also known to be a key factor associated with the progression of pancreatic cancer. Zhang *et al*., *Hepatobiliary Pancreat. Dis. Int.*, 2014, 13:81-86, incorporated by reference in its entirety.

[00353] In certain aspects, the antibody conjugates provided herein are useful for the treatment of one or more B cell malignancies. The B cell malignancy can be any B cell malignancy known to the practitioner of skill. These include B-cell malignancies described in the Revised European-American Lymphoma classification system (REAL) and those described in Harris *et al.*, 1994, Blood 84:1361-1392 and Armitage & Weisenburger, 1998, *J. Clin. Oncol.* 16:2780-2795, each incorporated by reference in its entirety. In certain embodiments, the B cell malignancy is selected from the group consisting of diffuse large B-cell lymphopma (DLBCL), follicular lymphoma, marginal zone B-cell lymphoma (MZL), mucosa-associated lymphatic tissue lymphoma (MALT), small lymphocytic lymphoma, mantle cell lymphoma (MCL). In certain embodiments, the DLBCL is selected from the group consisting of primary mediastinal (thymic) large B cell lymphoma, T cell/histiocyte-rich large B-cell lymphoma, primary cutaneous diffuse large B-cell lymphoma, leg type (primary cutaneous DLBCL, leg type), EBV positive diffuse large B-cell lymphoma of the elderly, diffuse large B-cell lymphoma associated with inflammation. In certain embodiments the B-cell malignancy is selected from the group consisting of Burkitt's lymphoma, lymphoplasmacytic lymphoma, Waldenström's macroglobulinemia, nodal marginal zone B cell lymphoma (NMZL), splenic marginal zone lymphoma (SMZL), intravascular large B-cell lymphoma, primary effusion lymphoma, lymphomatoid granulomatosis, primary central nervous system lymphoma, ALK-positive large B-cell lymphoma, plasmablastic lymphoma, large B-cell lymphoma arising in HHV8-associated

multicentric Castleman's disease, B-cell lymphoma (unclassifiable with features intermediate between diffuse large B-cell lymphoma and Burkitt lymphoma), and B-cell lymphoma (unclassifiable with features intermediate between diffuse large B-cell lymphoma and classical Hodgkin lymphoma). In certain embodiments, the malignancy is selected from Hodgkin's lymphoma, classic Hodgkin's lymphoma, and nodular lymphocyte predominant Hodgkin's lymphoma. In certain embodiments, the malignancy is non-Hodgkin's lymphoma.

[00354] In certain aspects, the antibody conjugates provided herein are useful for the treatment of one or more leukemias. In certain embodiments, the leukemia is acute leukemia. In certain embodiments, the leukemia is chronic leukemia. In certain embodiments, the leukemia is selected from the group consisting of acute lymphoblastic leukemia (ALL), chronic lymphocytic leukemia (CLL), acute myelogenous leukemia (AML), chronic myelogenous leukemia (CML), and hairy cell leukemia (HCL).

15. Diagnostic Applications

[00355] In some embodiments, the antibody conjugates provided herein are used in diagnostic applications. For example, an ant-CD74 antibody may be useful in assays for CD74 protein. In some aspects the antibody can be used to detect the expression of CD74 in various cells and tissues. These assays may be useful, for example, diagnosing cancer, infection and autoimmune disease.

[00356] In some diagnostic applications, the antibody 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 of the invention, the anti-CD74 antibody need not be labeled, and the presence thereof can be detected using a labeled antibody which specifically binds to the anti-CD74 antibody.

16. Affinity Purification Reagents

[00357] The antibody conjugates of the invention 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 is contacted with a sample containing the CD74 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 CD74 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 CD74 protein from the antibody.

17. Kits

[00358] In some embodiments, an antibody of the present invention can be provided in 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.

EXAMPLES

Example 1: Production and Purification of Anti-CD74 Antibodies

[00359] Antibodies were expressed in an Xpress CF™ reaction as described previously with the following modifications. The cell free extract for this work were created from an OmpT sensitive RF1 attenuated *E. coli* strain engineered to overexpress *E. coli* DsbC and FkpA as well as an orthogonal tRNA containing the CUA anti-codon for decoding the Amber Stop Codon. Extract was treated with 75 µM iodoacetamide for 45 min at RT (20°C) and added to a premix containing all other components, except for IgG heavy and light chain DNA. The final concentration in the protein synthesis reaction was 30%(v/v) cell extract, 2 mM para-azidomethylphenylalanine (pAMF) (RSP Amino Acids), 5uM engineered pAMF-specific amino-acyl tRNA synthetase (FRS variant), 2 mM GSSG, 8 mM magnesium glutamate, 10 mM ammonium glutamate, 130 mM potassium glutamate, 35 mM sodium pyruvate, 1.2 mM AMP, 0.86 mM each of GMP, UMP, and CMP, 2 mM amino acids (except 0.5 mM for Tyrosine and Phenylalanine), 4 mM sodium oxalate, 1 mM putrescine, 1.5 mM spermidine, 15 mM potassium phosphate, 100 nM T7 RNAP, 1 µg/mL antiCD74 light chain DNA, and 4 µg/mL antiCD74 heavy chain DNA. Site directed mutagenesis was used to introduce an amber stop codon (TAG) into the nucleotide sequence to encode for the pAMF non-natural amino acid at positions S7 and F404 (light and heavy chains respectively, kabat numbering). Cell free reactions were initiated by addition of plasmid DNA and incubated at 30°C for 16h in 100x10 mm petri dishes containing 10 mL.

[00360] The anti-CD74 cell free reactions were clarified by centrifugation at 10,000 rpm's for 30 minutes. The clarified supernatant was applied to Protein A MabSelect SuRe (GE Healthcare) with standard wash and low pH elution. Impurities such as aggregates were removed via preparative SEC (Sepax SRT-10C) equilibrated in 50 mM sodium phosphate, 200 mM arginine, pH 6.5. Final formulation of the sample was done in Dulbecco's Phosphate Buffered Saline (1x DPBS).

Example 2: Production of Anti-CD74 Antibodies with Non-natural Amino Acids

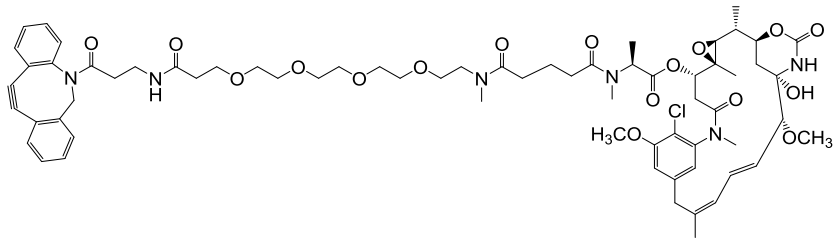
[00361] Antibodies were prepared having non-natural amino acids at positions heavy chain residues 404, 241, and 222, according to the EU number scheme, and at light chain residue 7, according to the Kabat or Chothia numbering scheme. One antibody comprised residue (56), above, at position 404, and four antibodies comprised residue (30), above, at each of postions 404, 241, 222 (heavy chain) and 7 (light chain). The starting heavy chain was according to SEQ ID NO:236, and the starting light chain was according to SEQ ID NO:256.

[00362] Each antibody was expressed at a total yield of at least 400 mg/L as shown in FIG.2A, and intact IgG were detected by SDS-PAGE as shown in FIG. 2B.

Example 3: Production of Antibody-PEG$_4$-Maytansine Conjugate

[00363] Purified anti-CD74 IgG containing modified amino acid residue 30 (*i.e.* para-azido-methyl-L-phenylalanine, or pAMF) at EU position 404 in its heavy chains was obtained according to Example 2. The anti-CD74 IgG was conjugated to a cytotoxic agent, maytansine, using a strained cyclooctyne reagent to yield **Conjugate A**.

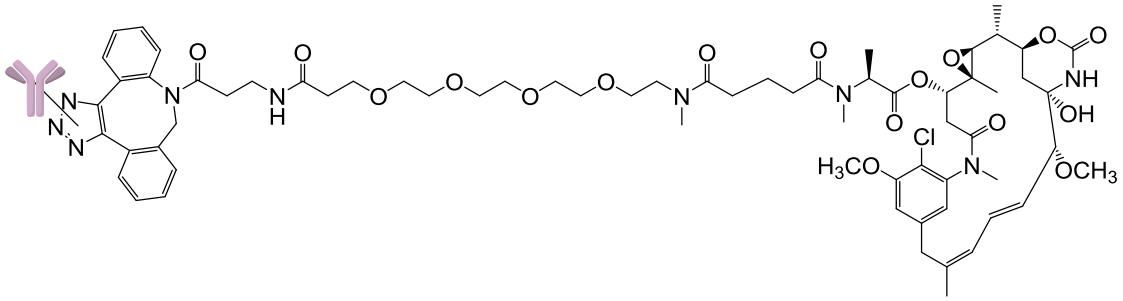
[00364] In brief, DBCO-PEG$_4$-maytansine (ACME Bioscience; Palo Alto, CA) according to the following:



was dissolved in DMSO to a final concentration of 5 mM. The compound was added to 1 mg/mL purified protein in PBS at a drug to antibody molar ratio of 12 to 1. The reaction mixture was incubated at RT (20°C) for 17 hours. Excess free drug was removed by Zeba plate (Thermo Scientific) equilibrated in PBS.

[00365] DAR analysis was done by MALDI-TOF (Bruker AutoFlex Speed). The conjugated protein was reduced for 10 min at 37°C with 10 mM TCEP in water and diluted to a final concentration of 50 µg/mL in 30% acetonitrile, 0.1% trifluoroacetic acid. Samples were combined 1:1 with S-DHB MALDI matrix (50 mg/mL in 50% acetonitrile, 0.1% trifluoroacetic acid) and 1 uL was applied to the MALDI target and dried under vacuum. Each MALDI spectra was accumulated for 5000 shots at full laser power in linear mode and the final DAR analysis was calculated by comparing the relative peak heights for conjugated and unconjugated masses for both the heavy and light chains.

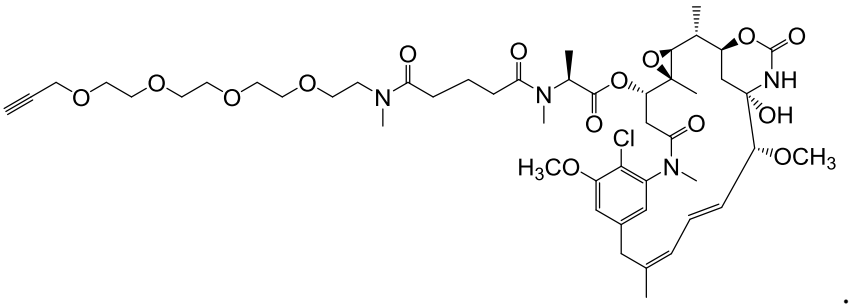
[00366] By peak intensity, MALDI-TOF showed a drug to antibody ratio (DAR) of 3.92. **Conjugate A**, as two regioisomers:



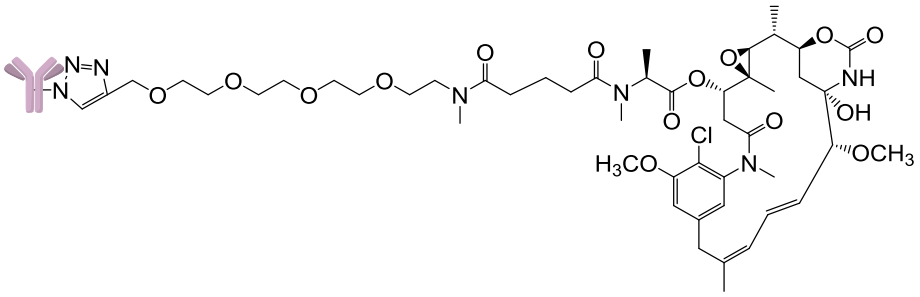
Example 4: Production of Antibody Conjugates

[00367] Purified anti-CD74 IgG containing modified amino acid residue 30 according to Example 2 was conjugated to maytansine, hemiasterlin, amanitin, MMAF, and MMAE linker-payload precursors to yield several anti-CD74-linker-payload conjugates designated **Conjugates B-F**.

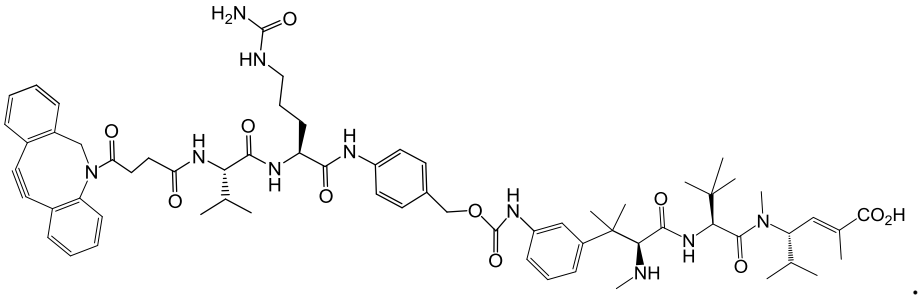
[00368] **Conjugate B** was prepared following a protocol consistent with Example 3 with the following PEG_4-maytansine precursor:



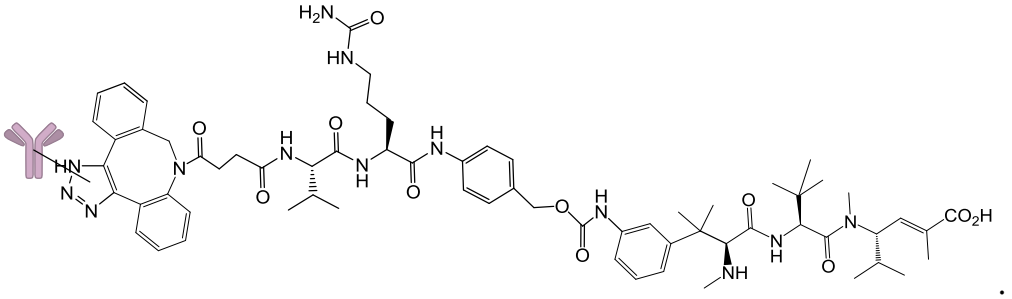
Conjugate B has the following structure, or a regioisomer thereof:



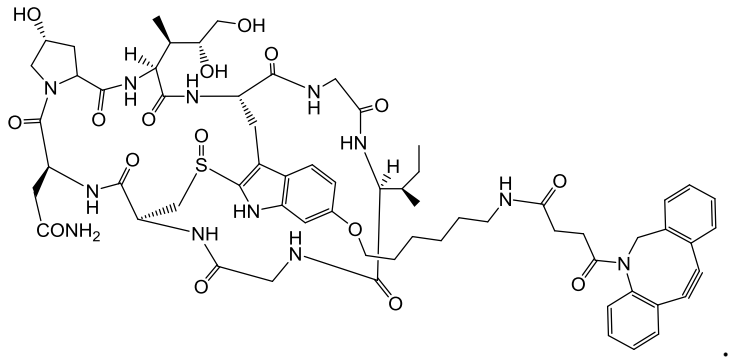
[00369] **Conjugate C** was prepared with the following val-cit-PAB-hemiasterlin precursor as described in U.S. application no. 62/110,390, filed January 30, 2015, entitled Hemiasterlin Derivatives for Conjugation and Therapy, incorporated by reference herein:



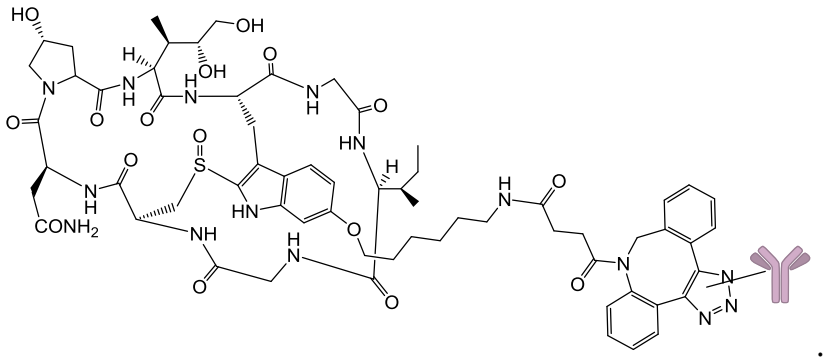
Conjugate C has the following structure, shown as two regioisomers:



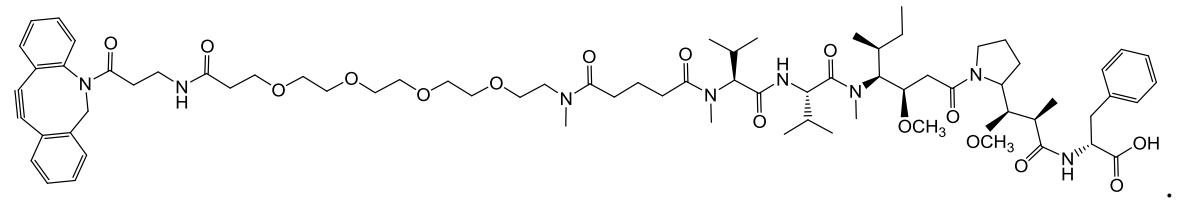
[00370] **Conjugate D** was prepared with the following linker-amanitin precursor:



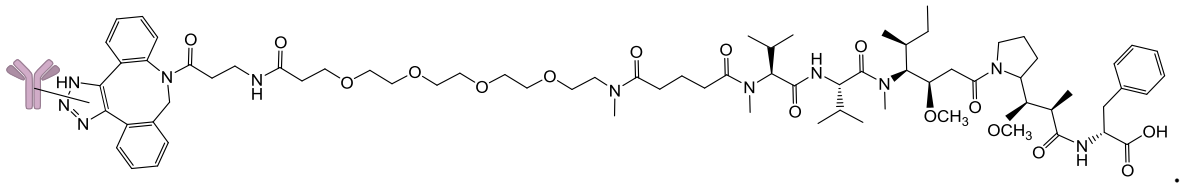
Conjugate D was prepared via O-alkylation of α-amanitin (BioTrend LLC, Destin, Florida) with *tert*-butyl (6-bromohexyl)carbamate, followed by cleavage of the Boc protecting group and acylation with DBCO succinate. **Conjugate D** has the following structure, shown as two regioisomers:



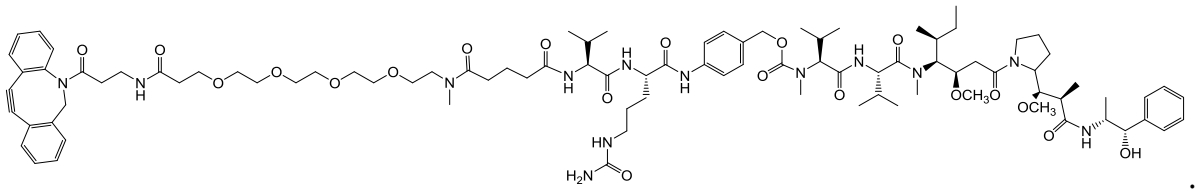
[00371] **Conjugate E** was prepared with the following PEG_4-MMAF precursor:



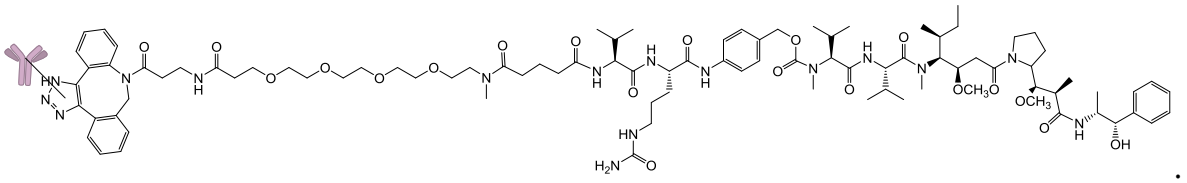
Conjugate E was prepared via acylation of MMAF (Doronina, S. *et al* 2003 *Nat Biotechnol* 21, 778-84) adapting the basic method outlined for **Conjugate A**. **Conjugate E** has the following structure, shown as two regioisomers:



[00372] **Conjugate F** was prepared with the following PEG_4-MMAE precursor:



Conjugate F was prepared by modification of the method described in Doronina, S. *et al* 2003 *Nat Biotechnol* 21, 778-84. **Conjugate F** has the following structure, shown as two regioisomers:



[00373] Expression of each conjugate was measured by absorbance at 280 nm, and the extent of conjugation was measured by MALDI-TOF. Control was the unconjugated antibody of Example 3. The data is summarized in the following table:

Conjugate	mg/mL (A280)	DAR	Buffer
Control	17.6		PBS
A	1.90	1.94	PBS +0.2 mM pAMF
B	2.10	1.96	PBS +0.2 mM pAMF
C	2.10	1.88	PBS +0.2 mM pAMF
D	2.04	1.95	PBS +0.2 mM pAMF
E	1.73	1.91	PBS +0.2 mM pAMF
F	1.79	1.73	PBS +0.2 mM pAMF

Example 5: Cell Binding and Cell Killing

[00374] **Conjugate A** was evaluated for the ability to bind and kill cells expressing CD74 by the methods below. Cell lines tested included B-lymphoma, multiple myeloma, and leukemia cells. Controls included unconjugated anti-CD74 antibody and free linker-drug (DIBCO-PEG_4-maytansine).

Cell Binding Assay

[00375] Cell lines were maintained in RPMI, high glucose (Cellgro-Mediatech; Manassas, VA) supplemented with 20% heat-inactivated fetal bovine serum (Hyclone; Thermo Scientific; Waltham, MA), 2mM glutamax (Invitrogen; Carlsbad, CA) and 1x Pencillin/streptomycin (Cellgro-Mediatech; Manassas, VA). Cells were harvested and re-suspended in FACS buffer (DPBS buffer supplemented with 1% bovine serum albumin). A total of 200,000 cells per well were incubated on ice with serial dilutions of anti-CD74 antibody without conjugation for 60 minutes. Cells were washed twice with ice-cold FACS buffer and incubated with 5ug/ml Alexa 647 labeled donkey anti-human IgG antibody (Jackson Immune-Research) on ice for another 60 mins. Unstained cells and cells stained with secondary antibody alone were used as controls. Samples were then washed twice using FACS buffer and analyzed using a BD FACS Canto system. Mean fluorescence intensities were fitted using non-linear regression analysis with one site specific binding equation on GraphPad Prism. Data was expressed as geometric mean fluorescent intensity vs. antibody concentration in nM.

Cell Killing Assay

[00376] Cytotoxicity effects of the free drug linkers and conjugates were measured with a cell proliferation assay. A total of 12500 cells in a volume of 25µl were seeded in a 384-well flat bottom white polystyrene plate on the day of assay. Free drug-linkers and conjugates were formulated at 2x starting concentration (1000nM for free drug linkers and 100nM for ADCs) in RPMI medium and filtered through MultiScreen HTS 96-Well Filter Plates (Millipore). Filter sterilized samples were serial diluted (1:3) under sterile conditions and added into treatment wells. Plates were cultured at 37°C in a CO_2 incubator for 72 hrs. For cell viability measurement, 30µl of Cell Titer-Glo® reagent (Promega Corp.) was added into each well, and plates processed as per product instructions. Relative luminescence was measured on an ENVISION® plate reader (Perkin-Elmer; Waltham, MA). Relative luminescence readings were converted to % viability using untreated cells as controls. Data was fitted with non-linear regression analysis, using log(inhibitor) vs. response, variable slope, 4 parameter fit equation using GraphPad Prism. Data was expressed as % relative cell viability vs. dose of free drug-linker or conjugate in nM.

[00377] Results for **Conjugate A** are summarized in the following Table 2:

Table 2. Results for **Conjugate A**

Diseases	Cell Lines Tested	Cell Binding		Cell Killing Activity			
		anti-CD74		Conjugate A		Free Linker-Drug	
		Bmax	Kd (nM)	Span (%)	IC50 (nM)	Span (%)	IC50 (nM)
B-Lymphoma	RPMI-6666 (HL)	3879	2.3	84	1.0	85	~ 13
	SU-DHL-6 (NHL)	1565	2.0	98	0.6	98	15.0
Multiple Myeloma	ARD (MM)	190	2.6	38	8.6	40	~ 34
	ARP-1 (MM)	341	2.8	74	9.0	77	17.0
	RPMI-8226 (MM)	119	3.6	NK	NK	NK	18.0
	OPM-2 (MM)	NB	NB	NK	NK	NK	24.0
Leukemia	BDCM (AML)	3059	4.5	74	4.6	76	13.0
	SUP-B15 (ALL)	680	3.5	64	3.0	65	11.0
	JVM-13 (CLL)	447	2.5	58	1.5	65	25.0
	K562 (CML)	NB	NB	NK	NK	NK	60.0

NK = no killing

NB = no binding

Example 6: Cell Binding and Cell Killing

[00378] Each of **Conjugates A-F** was evaluated for the ability to bind and kill cells expressing CD74. Cell lines tested included B-lymphoma, multiple myeloma, and leukemia cells. Controls included unconjugated anti-CD74 antibody. The results are summarized in the following Table 3:

Table 3. Results for **Conjugates A-F**

Disease	Cell Lines Tested	Cell Binding		Cell Killing Activity (Conjugate)											
		anti-CD74		A		B		C		D		E		F	
		Bmax	Kd (nM)	IC50 (nM)	Span (%)	IC50 (nM)	Span (%)	IC50 (nM)	Span (%)	IC50 (nM)	Span (%)	IC50 (nM)	Span (%)	IC50 (nM)	Span (%)
B-Lymphoma	RPMI-6666 (HL)	3879	2.3	1.0	85	1.3	89	0.9	84	4.3	69	1.5	80	0.7	82
	SU-DHL-6 (NHL)	1565	2.0	0.6	98	1.3	98	0.3	97	0.7	99	0.5	98	0.3	97
Multiple Myeloma	ARD (MM)	190	2.6	8.6	40	31.0	53	26.0	74	7.1	25	18.0	20	16.0	73
	ARP-1 (MM)	341	2.8	9.0	77	16.0	82	7.6	87	1.7	91	6.0	71	4.2	93
	RPMI-8226 (MM)	119	3.6	NK	NK	51.0	41	17.0	43	NK	NK	NK	NK	20.0	31
	OPM-2 (MM)	NB	NB	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK
Leukemia	BDCM (AML)	3059	4.5	4.6	76	15.0	95	1.1	89	2.2	84	3.3	78	1.4	94
	SUP-B15 (ALL)	680	3.5	3.0	65	5.7	72	2.8	68	2.4	74	2.5	58	3.7	65
	JVM-13 (CLL)	447	2.5	1.5	65	2.0	69	0.9	54	6.8	73	7.8	47	5.0	59
	K562 (CML)	NB	NB	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK	NK

Example 7: Evaluation of *in vivo* anti-tumor activity in a mouse multiple myeloma model

[00379] A study was carried out to investigate the *in vivo* anti-tumor efficacy of an anti-CD74 antibody-drug conjugate in a human multiple myeloma model (disseminated). **Conjugate A** (Example 3) was evaluated in animals inoculated with ARP-1 multiple myeloma cells. ARP-1 (and isogenic cell lines ARD-1 and CAG) were established from bone marrow aspirates of a patient with multiple myeloma patient (Kwong, K. Characterization of an Isogenic Model System for KDM6A/UTX Loss in Multiple Myeloma. (2013), incorporated herein by reference in its entirety).

[00380] *Protocol.* Female CB17 SCID (severe combined immunodeficiency) were inoculated with ARP-1 cells via tail vein injection. Prior to tumor cell inoculation, animals were pre-treated with 0.4 mg/mouse Fludarabine and 2 mg/mouse Cyclophosphamide (intraperitoneal injection). Randomization and treatment was initiated 14 days post tumor inoculation. Treatment groups are outlined in Table 4. All test articles were formulated in 10 mM citrate pH 6.0, 10% sucrose and diluted with PBS. Animals were administered with the designated test article four times every 7 days (q7dx4) by intravenous injection. Untreated animals did not receive chemo pre-treatment and were not inoculated with ARP-1 cells. Body weights were monitored twice per week. When vehicle control animals reached the study endpoint (euthanized due to large, palpable internal tumors), three animals from the Group 2 and Group 3 were euthanized for necropsy and bone marrow harvest (from tibia and femur) for flow cytometry. The remaining animals in treated groups were monitored for up to 3 months after start of treatment.

Table 4. Treatment groups

Group	Treatment	DAR	Dose (mg/kg)	Route	N
1	Vehicle (PBS)	-	-	IV	10
2	Conjugate A	1.97	3	IV	10
3	Untreated	-	-	-	3

[00381] Bone marrow cells from mouse femur and tibia were pooled and assessed for CD138 expression using the Alexa Fluor 647 mouse anti-human CD138 clone MI15 (BD Biosciences #562097) according to the manufacturer's protocol. Direct immunofluorescence

flow cytometric analysis was performed using an LSRII flow cytometer and FACS Diva Software. Data was analyzed using Flowjo (Tree Star, Inc., Ashland, OR).

[00382] Body weight, tissue volume, or tissue weights at study endpoint, were analyzed using a one-way analysis of variance (ANOVA) with Dunnett's multiple comparison test. A probability of less than 5% ($p<0.05$) was considered as significant.

[00383] *Results.* Multiple myeloma cells, ARP-1, were inoculated intravenously into CB17 SCID mice and treated with 3 mg/kg **Conjugate A** (q7dx4). Study endpoint was characterized by noteworthy body weight change, formation of large palpable internal tumors, and moribundity. FIG. 3A shows steady weight gain (increase in BWC) in vehicle control animals starting on approximately day 25 post tumor inoculation, and subsequent development of distended abdomens by day 35. On day 49, vehicle control animals were euthanized based on clinical endpoints and palpable tumors in the abdomen. No meaningful change in body weight (FIG. 3A), enlarged abdomen, or signs of distress were observed in groups treated with **Conjugate A**. The BWC on the last recorded day for vehicle control group (day 46) was trending toward significant (**Conjugate A**, $p = 0.0551$) compared to vehicle control (FIG. 3B).

[00384] Gross pathological analysis of vehicle control animals revealed large tumor masses in and around the ovaries and/or kidneys (FIGS. 4A, 4B). Formation of internal tumors likely accounts for distended abdomens and increase in body weight observed in vehicle control group. A subset of animals from groups treated with **Conjugate A** (n=3 per group) were harvested for anatomical examination and flow cytometry. FIGS. 4A and 4B show that kidney and ovaries from both groups that received CD74 ADC treatment looked phenotypically normal, and had comparable tissue weights to untreated age-matched control animals (which did not receive cells or treatment). Tumor burden was also assessed by measuring the percentage of human CD138 positive ARP-1 myeloma cells present in the bone marrow (femur and tibia). CD138 is a specific surface antigen for multiple myeloma and plasma cells in the bone marrow (Chilosi, M. *et al.* "CD138/syndecan-1: A useful immunohistochemical marker of normal and neoplastic plasma cells on routine trephine bone marrow biopsies." *Mod. Pathol. Off. J. U. S. Can. Acad. Pathol. Inc* **12,** 1101−1106 (1999), incorporated herein by reference in its entirety). The vehicle control group showed high tumor burden, while treatment with **Conjugate A** substantially inhibited ARP-1 growth in the bone marrow (FIGS. 5A, 5B). Bone marrow isolated from untreated

age-matched controls looked similar to the **Conjugate A**-treated group. Cumulatively, these results demonstrate that **Conjugate A** abrogated tumor burden.

[00385] Remaining animals in the **Conjugate A** treatment group (n=6-7 per group) continued to be monitored for disease progression after the first round of treatment. Treatment with **Conjugate A** similarly delayed weight gain for approximately 20 days, followed by a steady increase in weight starting at approximately day 70 or 35 days after the last dose. By day 82 (45 days after last treatment), some animals developed distended abdomens and large tumor mass around ovaries and/or kidneys. A subset of animals from the treatment group (n=3) received a second round of **Conjugate A** treatment starting on day 82. Apparent stabilization of weight gain was observed, suggesting prevention of disease progression.

[00386] Results from this study show that **Conjugate A** attenuated body weight increase and reduced tumor burden (in bone marrow and for internal tumors) with similar potency and duration of response in animals inoculated with ARP-1 multiple myeloma cells.

Example 8: Further studies of *in vivo* anti-tumor activity in a mouse multiple myeloma model

[00387] A study was carried out to investigate the *in vivo* anti-tumor efficacy of an anti-CD74 antibody-drug conjugate in a human multiple myeloma model (disseminated). **Conjugate A** (Example 3) was evaluated in animals inoculated with MM.1S multiple myeloma cells.

[00388] *Protocol.* Female NOD SCID gamma (NSG) mice 8 weeks of age were inoculated with multiple myeloma MM.1S cells into the tail vein. Randomization by body weight and start of treatment was initiated 11 days post tumor inoculation. Treatment groups are outlined in Table 5. Test articles were formulated in 10 mM citrate, pH 6.0, 10% sucrose and diluted in PBS for administration. Animals were administered with **Conjugate A** every 7 days for 3 weeks (q7dx3) by intravenous (IV) injection. For each group, a subset of animals was used for bone marrow harvest and analysis (n=3) and the remaining animals (n=5) were monitored for survival. Survival endpoint was characterized by >20% body weight loss and clinical signs including lethargy, hind limb paralysis or moribundity. On day 32 (3 weeks after start of treatment), 3 animals from each group were randomly chosen to be euthanized for bone marrow harvest (pooled from tibia and femur) for flow cytometry. On day 129 (~4 months after start of treatment), all surviving animals were euthanized and bone marrow was harvested (pooled from tibia and femur) for flow cytometry.

Table 5. List of Treatment Groups

Group	Treatment	Dose (mg/kg)	Dosing frequency	Route	N
1	Vehicle control	na	q7dx3	IV	8
2	**Conjugate A**	3	q7dx3	IV	8
3	**Conjugate A**	10	q7dx3	IV	8

[00389] Body weights were monitored at least twice per week for up to 129 days. All graphs are presented as mean ± standard error of the mean (SEM) versus days post-treatment start.

[00390] Percent body weight change (BWC) is calculated relative to weight on the day of treatment. Substantial toxicity was defined as a >20% decrease in animal weight, at which point affected animals are euthanized.

[00391] Bone marrow cells from mouse femur and tibia were pooled and assessed for human CD138 expression using the Alexa Fluor 647 mouse anti-human CD138 clone MI15 (BD Biosciences #562097) according to the manufacturer's protocol. CD138 is a specific surface antigen for MM and plasma cells in the bone marrow (Chilosi, M. *et al.* (1999), *supra*). Direct immunofluorescence flow cytometric analysis was performed using an LSRII flow cytometer and FACS Diva Software. Data was analyzed using Flowjo (Tree Star, Inc., Ashland, OR).

[00392] Human CD138+ cells in the bone marrow on day 32 was analyzed using one-way analysis of variance (ANOVA) with a Dunnett's adjustment at 0.05 significance level. Data from MM.1s positive control were excluded from analysis. A probability of less than 5% ($p<0.05$) was considered as significant.

[00393] *Results.* Multiple myeloma MM.1S cells were inoculated intravenously into NSG mice and treated with vehicle, 3 mg/kg or 10 mg/kg **Conjugate A** (q7dx3) starting on day 11 post-tumor inoculation. The survival study endpoint criteria included body weight change greater than 20% and clinical signs of moribundity. FIG. 6A shows body weight loss in vehicle control animals starting approximately on day 28 post tumor inoculation. Body weight loss was accompanied by hunched posture, hind-limb paralysis and severe lethargy. The mean survival for the vehicle group was 35 days, while treatment with 3 or 10 mg/kg **Conjugate A** resulted in 100% survival with all animals showing no sign of disease on day 129 post inoculation (FIG. 6B).

[00394] On day 32, tumor burden was assessed by measuring the percentage of hCD138+ myeloma cells in the bone marrow. FIG. 6C shows that both doses of **Conjugate A** significantly reduced tumor burden compared to high tumor burden (~50%) in the vehicle control group. After approximately 4 months (Day 129), no hCD138+ cells were detected in the bone marrow of animals treated with 3 and 10 mg/kg **Conjugate A** (FIG. 6D). Both data sets on day 32 and day 129 included bone marrow from non-inoculated negative control animals (no cells or treatment) and MM.1S cells as a positive control. As expected, the non-inoculated group and MM.1S cells had low and high hCD138+, respectively.

[00395] Results from this study show that 3 or 10 mg/kg **Conjugate A** (q7dx3) eradicated disease based on 100% survival and absence of hCD138+ in the bone marrow approximately 4 months after initiation of treatment in the disseminated MM.1S model.

Example 9: Evaluation of *in vivo* anti-tumor activity in a mouse non-Hodgkin's lymphoma (NHL) model

[00396] A study was carried out to investigate the *in vivo* anti-tumor efficacy of an anti-CD74 antibody-drug conjugate in a human non-Hodgkin′s lymphoma ("NHL") model. **Conjugate A** (Example 3) was evaluated in the animals bearing established subcutaneous SU-DHL-6 NHL tumors (diffuse large B-cell lymphoma). Treatment with **Conjugate A** significantly delayed growth of NHL tumors SU-DHL-6.

[00397] *Protocol.* Female CB17 SCID mice 9 weeks of age were anesthetized with isoflurane and implanted subcutaneously into the right flank with with a 1:1 mixture of SU-DHL-6 cells and matrigel. Randomization, enrollment into treatment groups, and start of treatment was initiated approximately 14 days post implantation. Treatment groups are outlined in Table 6. All test articles were formulated in 10 mM citrate pH 6.0, 9% sucrose and diluted in PBS. Body weight and tumor size were monitored twice per week until mean of control treated tumors were >1000 mm^3 or end of study. Animal body weights included the tumor weight. Percent body weight change (BWC) is calculated relative to weight on the day of treatment. No samples were collected at the end of study.

Table 6. Treatment groups

Group	Treatment	DAR	Dose (mg/kg)	Dosing frequency	Route	N
1	Vehicle	-	-	q7dx3	IV	7
2	**Conjugate A**	1.97	10	q7dx3	IV	7

[00398] Tumor size on day 18 post-tumor implantation (last day control animals were on study) was analyzed using a one-way analysis of variance (ANOVA) with Dunnett's multiple comparison test. A probability of less than 5% ($p<0.05$) was considered as significant.

[00399] *Results.* In this study, NHL cells SU-DHL-6 were implanted subcutaneously into CB17 SCID mice and treated with 10 mg/kg **Conjugate A**. Efficacy results from a repeat dosing regimen (q7dx3) is presented in FIG. 7A and FIG. 7B. Treatment with **Conjugate A** (q7dx3) significantly inhibited SU-DHL-6 tumor growth, and resulted in ~80% tumor growth inhibition compared to control on day 18 (**** $p<0.0001$, Table 6). It was noted that there was some continued growth of SU-DHL-6 tumors in the presence of treatment with **Conjugate A**. In addition, treatment with **Conjugate A** was well tolerated and did not exhibit any toxicity based on the absence of significant effects on animal body weights (FIG. 8).

Table 6. Statistical comparison of tumor size on day 18 versus vehicle control

Group compared to vehicle (1246 mm^3 on day 21)	Average final tumor size on day 18	Significant?	Adjusted p value
10 mg/kg **Conjugate A**	291 mm^3	Yes	<0.0001

[00400] Results from this study show that **Conjugate A** is significantly efficacious in slowing SU-DHL-6 tumor growth.

Example 10: Sequences

[00401] Table 7 provides sequences referred to herein.

Table 7. Sequences.

SEQ ID NO	Molecule	Region	Scheme	Sequence
1	1251-B08	CDR-H1	Chothia	GFNFSDY
2	1193-C08	CDR-H1	Chothia	GFTFNNN
3	1193-E06b	CDR-H1	Chothia	GFTFNNT
4	1193-H04b	CDR-H1	Chothia	GFTFTSS
5	1198-A01	CDR-H1	Chothia	GFTFSDY
6	1198-B10	CDR-H1	Chothia	GFNISGS
7	1198-D03	CDR-H1	Chothia	GFNINNY
8	1198-D04	CDR-H1	Chothia	GFNINNY
9	1251-A02	CDR-H1	Chothia	GFAFSDH
10	1251-A03	CDR-H1	Chothia	GFAFSDH
11	1251-A06	CDR-H1	Chothia	GFDFSSY
12	1193-B06	CDR-H1	Chothia	GFTFTGN
13	1251-B09	CDR-H1	Chothia	GFNFSDY
14	1251-B10	CDR-H1	Chothia	GFNFSSH
15	1251-C03	CDR-H1	Chothia	GFNFSSY
16	1251-D02	CDR-H1	Chothia	GFSFASH
17	1251-D06	CDR-H1	Chothia	GFSFGSY
18	1251-D09	CDR-H1	Chothia	GFSFSSY
19	1251-E06	CDR-H1	Chothia	GFTFDSY
20	1251-F06	CDR-H1	Chothia	GFTFSSF
21	1251-F07	CDR-H1	Chothia	GFTFSSH
22	1251-G02	CDR-H1	Chothia	GFTFSSY
23	1445-A03	CDR-H1	Chothia	GFNISGY
24	1445-B09	CDR-H1	Chothia	GFNITGT
25	1447-D11	CDR-H1	Chothia	GFTFNNT
26	1447-E08	CDR-H1	Chothia	GFTFNDT
27	1447-F11	CDR-H1	Chothia	GFTFDNT
28	1447-G01	CDR-H1	Chothia	GFTFNTS
29	VH11-[19]	CDR-H1	Chothia	GFTFSSY
30	VH5-[7]	CDR-H1	Chothia	GFTFSSY
31	VH6-[11]	CDR-H1	Chothia	GFTFSSY

SEQ ID NO	Molecule	Region	Scheme	Sequence
32	VH8-[15]	CDR-H1	Chothia	GFTFSSY
33	1251-B08	CDR-H1	Kabat	DYGMH
34	1193-C08	CDR-H1	Kabat	NNWMS
35	1193-E06b	CDR-H1	Kabat	NTDMS
36	1193-H04b	CDR-H1	Kabat	SSWMS
37	1198-A01	CDR-H1	Kabat	DYDMS
38	1198-B10	CDR-H1	Kabat	GSWIH
39	1198-D03	CDR-H1	Kabat	NYDIH
40	1198-D04	CDR-H1	Kabat	NYDIH
41	1251-A02	CDR-H1	Kabat	DHGMH
42	1251-A03	CDR-H1	Kabat	DHGMH
43	1251-A06	CDR-H1	Kabat	SYGMH
44	1193-B06	CDR-H1	Kabat	GNWMS
45	1251-B09	CDR-H1	Kabat	DYGMH
46	1251-B10	CDR-H1	Kabat	SHGMH
47	1251-C03	CDR-H1	Kabat	SYGMH
48	1251-D02	CDR-H1	Kabat	SHGMH
49	1251-D06	CDR-H1	Kabat	SYGMH
50	1251-D09	CDR-H1	Kabat	SYGMH
51	1251-E06	CDR-H1	Kabat	SYGMH
52	1251-F06	CDR-H1	Kabat	SFGMH
53	1251-F07	CDR-H1	Kabat	SHGMH
54	1251-G02	CDR-H1	Kabat	SYGMH
55	1445-A03	CDR-H1	Kabat	GYYIH
56	1445-B09	CDR-H1	Kabat	GTGIH
57	1447-D11	CDR-H1	Kabat	NTDMS
58	1447-E08	CDR-H1	Kabat	DTDMS
59	1447-F11	CDR-H1	Kabat	NTDMS
60	1447-G01	CDR-H1	Kabat	TSDMS
61	VH11-[19]	CDR-H1	Kabat	SYGMH
62	VH5-[7]	CDR-H1	Kabat	SYAMH
63	VH6-[11]	CDR-H1	Kabat	SYAMH

SEQ ID NO	Molecule	Region	Scheme	Sequence
64	VH8-[15]	CDR-H1	Kabat	SYAMH
65	1251-B08	CDR-H2	Chothia	WYDGSI
66	1193-C08	CDR-H2	Chothia	NGDDGY
67	1193-E06b	CDR-H2	Chothia	NGSGGA
68	1193-H04b	CDR-H2	Chothia	NGYNGI
69	1198-A01	CDR-H2	Chothia	AQDGSY
70	1198-B10	CDR-H2	Chothia	YPDDGD
71	1198-D03	CDR-H2	Chothia	DPYNGA
72	1198-D04	CDR-H2	Chothia	DPYNGT
73	1251-A02	CDR-H2	Chothia	WYDGSH
74	1251-A03	CDR-H2	Chothia	WYDGSH
75	1251-A06	CDR-H2	Chothia	WDDGSD
76	1193-B06	CDR-H2	Chothia	YGTSGA
77	1251-B09	CDR-H2	Chothia	WYDGSR
78	1251-B10	CDR-H2	Chothia	WHDGSD
79	1251-C03	CDR-H2	Chothia	WYDGSI
80	1251-D02	CDR-H2	Chothia	WDDGSD
81	1251-D06	CDR-H2	Chothia	WYDGSK
82	1251-D09	CDR-H2	Chothia	WYDASI
83	1251-E06	CDR-H2	Chothia	WYDGSN
84	1251-F06	CDR-H2	Chothia	WYDGSN
85	1251-F07	CDR-H2	Chothia	WDDGSN
86	1251-G02	CDR-H2	Chothia	WHDGSK
87	1445-A03	CDR-H2	Chothia	SPTGGY
88	1445-B09	CDR-H2	Chothia	TPYNGT
89	1447-D11	CDR-H2	Chothia	NGSGGS
90	1447-E08	CDR-H2	Chothia	NGAGGA
91	1447-F11	CDR-H2	Chothia	NGSGGV
92	1447-G01	CDR-H2	Chothia	NGSGGA
93	VH11-[19]	CDR-H2	Chothia	WYDGSN
94	VH5-[7]	CDR-H2	Chothia	SYDGSN
95	VH6-[11]	CDR-H2	Chothia	SYDGSI

SEQ ID NO	Molecule	Region	Scheme	Sequence
96	VH8-[15]	CDR-H2	Chothia	SYDGSN
97	1251-B08	CDR-H2	Kabat	VIWYDGSISYYADSVKG
98	1193-C08	CDR-H2	Kabat	IINGDDGYTYYADRVKG
99	1193-E06b	CDR-H2	Kabat	IINGSGGATNYADSVKG
100	1193-H04b	CDR-H2	Kabat	IINGYNGITYYADSVKG
101	1198-A01	CDR-H2	Kabat	FIAQDGSYKYYVDSVKG
102	1198-B10	CDR-H2	Kabat	YIYPDDGDTYYADSVKG
103	1198-D03	CDR-H2	Kabat	NIDPYNGATYYADSVKG
104	1198-D04	CDR-H2	Kabat	NIDPYNGTTYYADSVKG
105	1251-A02	CDR-H2	Kabat	VIWYDGSHKIYADSVKG
106	1251-A03	CDR-H2	Kabat	VIWYDGSHKIYADSVKG
107	1251-A06	CDR-H2	Kabat	VIWDDGSDRYYADSVKG
108	1193-B06	CDR-H2	Kabat	IIYGTSGATYYADSVKG
109	1251-B09	CDR-H2	Kabat	VTWYDGSREYYADSVKG
110	1251-B10	CDR-H2	Kabat	VIWHDGSDKYYADSVKG
111	1251-C03	CDR-H2	Kabat	VIWYDGSIKNYADSVKG
112	1251-D02	CDR-H2	Kabat	VIWDDGSDRYYADSVKG
113	1251-D06	CDR-H2	Kabat	VVWYDGSKTIYADSVKG
114	1251-D09	CDR-H2	Kabat	VIWYDASIRKYAGSVKG
115	1251-E06	CDR-H2	Kabat	VIWYDGSNKVYADSVKG
116	1251-F06	CDR-H2	Kabat	VIWYDGSNEYYADSVKG
117	1251-F07	CDR-H2	Kabat	VIWDDGSNEVYADSVKG
118	1251-G02	CDR-H2	Kabat	VIWHDGSKDYYADSVKG
119	1445-A03	CDR-H2	Kabat	EISPTGGYTYYADSVKG
120	1445-B09	CDR-H2	Kabat	IITPYNGTTNYADSVKG
121	1447-D11	CDR-H2	Kabat	VINGSGGSSNYADSVKG
122	1447-E08	CDR-H2	Kabat	MINGAGGASFYADSVRG
123	1447-F11	CDR-H2	Kabat	IINGSGGVTNYADSVRG
124	1447-G01	CDR-H2	Kabat	IINGSGGATNYADSVKG
125	VH11-[19]	CDR-H2	Kabat	VIWYDGSNKYYADSVKG
126	VH5-[7]	CDR-H2	Kabat	VISYDGSNKYYADSVKG
127	VH6-[11]	CDR-H2	Kabat	VISYDGSIKYYADSVKG

SEQ ID NO	Molecule	Region	Scheme	Sequence
128	VH8-[15]	CDR-H2	Kabat	VISYDGSNKYYADSVKG
129	1251-B08	CDR-H3	K/C	GGTVEHGAVYGTDV
130	1193-C08	CDR-H3	K/C	VALGRPRRFDY
131	1193-E06b	CDR-H3	K/C	FENEWEVSMDY
132	1193-H04b	CDR-H3	K/C	PSAPGARRFDY
133	1198-A01	CDR-H3	K/C	SKLFRAGQFDY
134	1198-B10	CDR-H3	K/C	EGSHNLDKMDY
135	1198-D03	CDR-H3	K/C	VLWGFWAPFDY
136	1198-D04	CDR-H3	K/C	VPWGFWAPFDY
137	1251-A02	CDR-H3	K/C	GGSLAGGAVYGTDV
138	1251-A03	CDR-H3	K/C	GGSLAGGAVYGTDV
139	1251-A06	CDR-H3	K/C	GGTRVLGAIHGTDV
140	1193-B06	CDR-H3	K/C	PSMSGSRGFDY
141	1251-B09	CDR-H3	K/C	GGTLVHGALYGNDV
142	1251-B10	CDR-H3	K/C	GGTRVLGAVYGLDV
143	1251-C03	CDR-H3	K/C	GGALMRGEFSGHDV
144	1251-D02	CDR-H3	K/C	GGTRVLGAIHGTDV
145	1251-D06	CDR-H3	K/C	GGTLVRGAVYGLDV
146	1251-D09	CDR-H3	K/C	GGTVERGAIYGTDV
147	1251-E06	CDR-H3	K/C	GGMVGQGAMFGLDV
148	1251-F06	CDR-H3	K/C	GGSLVTRGVYGLDV
149	1251-F07	CDR-H3	K/C	GGTRIRGLRYGTDV
150	1251-G02	CDR-H3	K/C	GGQLDHGAIYGLDV
151	1445-A03	CDR-H3	K/C	EHGLVYGQPMDY
152	1445-B09	CDR-H3	K/C	GGYGYYYPPFDY
153	1447-D11	CDR-H3	K/C	YETEWEVSLDY
154	1447-E08	CDR-H3	K/C	FENQWEVTFDY
155	1447-F11	CDR-H3	K/C	YESEWEVSLDY
156	1447-G01	CDR-H3	K/C	YENEMEVSMDY
157	VH11-[19]	CDR-H3	K/C	GGTLVRGAMYGTDV
158	VH5-[7]	CDR-H3	K/C	GRYYGSGSYSSYFDY
159	VH6-[11]	CDR-H3	K/C	GREITSQNIVILLDY

SEQ ID NO	Molecule	Region	Scheme	Sequence
160	VH8-[15]	CDR-H3	K/C	GREITSQNIVILLDY
161	1337-A09	CDR-L1	K/C	RASQGIGSWLA
162	1193-C08	CDR-L1	K/C	RASQSVSSNYLA
163	1193-E06b	CDR-L1	K/C	RASQSVSSSYLA
164	1193-H04b	CDR-L1	K/C	RASQSVSSSYLA
165	1275-C10	CDR-L1	K/C	RASQGVSSWLA
166	1275-D01	CDR-L1	K/C	RASQGIGRWLA
167	1275-D10	CDR-L1	K/C	RASQGVFSWLA
168	1275-G02	CDR-L1	K/C	RASQGLGSFLA
169	1337-A04	CDR-L1	K/C	RASQDIGRWVA
170	1337-A05	CDR-L1	K/C	RASQGIGRWVA
171	1337-A06	CDR-L1	K/C	RASQDIGSWVA
172	1337-A07	CDR-L1	K/C	RASQGISSWVA
173	1337-A08	CDR-L1	K/C	RASQDIGSWVA
174	1193-B06	CDR-L1	K/C	RAGQSVSSSYLA
175	1337-A10	CDR-L1	K/C	RASQGISSWVA
176	1447-D11	CDR-L1	K/C	RASQSVSSSYLA
177	1447-E08	CDR-L1	K/C	RASQRVAGIDLS
178	1447-F11	CDR-L1	K/C	RASQSVYRSYLA
179	1447-G01	CDR-L1	K/C	RASQSVSSRELG
180	VL-5[23] & VL6-[26]	CDR-L1	K/C	RASQGISSWLA
181	1337-A09	CDR-L2	K/C	AADRLQS
182	1193-C08	CDR-L2	K/C	GASSRAT
183	1193-E06b	CDR-L2	K/C	GASSRAT
184	1193-H04b	CDR-L2	K/C	GASSRAT
185	1275-C10	CDR-L2	K/C	SARYLQS
186	1275-D01	CDR-L2	K/C	GRSSLQS
187	1275-D10	CDR-L2	K/C	NATQLQS
188	1275-G02	CDR-L2	K/C	LGNLLQI
189	1337-A04	CDR-L2	K/C	GASSLQS
190	1337-A05	CDR-L2	K/C	GADRLQS
191	1337-A06	CDR-L2	K/C	GADRLQS

SEQ ID NO	Molecule	Region	Scheme	Sequence
192	1337-A07	CDR-L2	K/C	GASRLQS
193	1337-A08	CDR-L2	K/C	ASDSLQS
194	1193-B06	CDR-L2	K/C	GASSRAT
195	1337-A10	CDR-L2	K/C	GSSRLQS
196	1447-D11	CDR-L2	K/C	GASSRAT
197	1447-E08	CDR-L2	K/C	GASSRAT
198	1447-F11	CDR-L2	K/C	GASSRAT
199	1447-G01	CDR-L2	K/C	GASSRAT
200	VL-5[23] & VL6-[26]	CDR-L2	K/C	AASSLQS
201	1337-A09	CDR-L3	K/C	QQYHTYPLT
202	1193-C08	CDR-L3	K/C	QQHYTTPPT
203	1193-E06b	CDR-L3	K/C	QQHYTTPPT
204	1193-H04b	CDR-L3	K/C	QQHYTTPPT
205	1275-C10	CDR-L3	K/C	QQYNLYPLT
206	1275-D01	CDR-L3	K/C	QQYNIYPLT
207	1275-D10	CDR-L3	K/C	QQYYYYPLT
208	1275-G02	CDR-L3	K/C	QQYNAYPLT
209	1337-A04	CDR-L3	K/C	QQYNTYPLT
210	1337-A05	CDR-L3	K/C	QQYNSYPLT
211	1337-A06	CDR-L3	K/C	QQYNSYPLT
212	1337-A07	CDR-L3	K/C	QQYHTYPLT
213	1337-A08	CDR-L3	K/C	QQYNSYPLT
214	1193-B06	CDR-L3	K/C	QQHYTTPPT
215	1337-A10	CDR-L3	K/C	QQYNTYPLT
216	1447-D11	CDR-L3	K/C	QHNQPTPPT
217	1447-E08	CDR-L3	K/C	QQHNTTPPT
218	1447-F11	CDR-L3	K/C	QQHQTAPPT
219	1447-G01	CDR-L3	K/C	QQQCSWPPT
220	VL-5[23] & VL6-[26]	CDR-L3	K/C	QQYNSYPLT

SEQ ID NO	Molecule	Region	Scheme	Sequence
221	1193-B06	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF TGNWMSWVRQAPGKGLEWVGIIYGTSGAT YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKPSMSGSRGFDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRAGQSVSSSYLAWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQQHYTTPP TFGQGTKVEIK
222	1193-C08	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNNWMSWVRQAPGKGLEWVGIINGDDGYT YYADRVKGRFTIIRDNSKNTLYLQMNSLR AEDTAVYYCAKVALGRPRRFDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLTPGERATLSCRASQSVSSNYLAWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAMYYCQQHYTTPP TFGQGTKVEIK
223	1193-E06b	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNTDMSWVRQAPGKGLEWVGIINGSGGAT NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKFENEWEVSMDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRASQSVSSSYLAWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQQHYTTPP TFGQGTKVEIK
224	1193-H04b	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF TSSWMSWVRQAPGKGLEWVGIINGYNGIT YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKPSAPGARRFDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGEATLSCRASQSVSSSYLAWYQQR PGQAPRLLIYGASSRATGIPDRFSGSGSG TDFTLTISRLEPEDFAVYYCQQHYTTPPT FGQGTKVEIK
225	1447-D11	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNTDMSWVRQAPGKGLEWVGVINGSGGSS NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYETEWEVSLDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRASQSVSSSYLAWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQHNQPTPP TFGQGTKVEIK

SEQ ID NO	Molecule	Region	Scheme	Sequence
226	1447-E08	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF NDTDMSWVRQAPGKGLEWVGMINGAGGAS FYADSVRGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKFENQWEVTFDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRASQRVAGIDLSWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQQHNTTPP TFGQGTKVEIK
227	1447-F11	scFv		EVQLLESGGGLVQTGGSLRLSCAASGFTF DNTDMSWVRQAPGKGLEWVGIINGSGGVT NYADSVRGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYESEWEVSLDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRASQSVYRSYLAWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQQHQTAPP TFGQGTKVEIK
228	1447-G01	scFv		EVQLLESGGGLVQPGGSLRLSCAASGFTF NTSDMSWVRQAPGKGLEWVGIINGSGGAT NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYENEMEVSMDYWGQGTLV TVSSGGGGSGGGGSGGGGSEIVLTQSPGT LSLSPGERATLSCRASQSVSSRELGWYQQ KPGQAPRLLIYGASSRATGIPDRFSGSGS GTDFTLTISRLEPEDFAVYYCQQQCSWPP TFGQGTKVEIK
229	1251-B08-g_1337-A09-g scFv-Fc	scFv-Fc		QVQLVESGGGVVQPGRSLRLSCAASGFNF SDYGMHWVRQAPGKGLEWVAVIWYDGSIS YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTVEHGAVYGTDVWGQG TTVTVSSGGGGSGGGGSGGGGSDIQMTQS PSSVSASVGDRVTITCRASQGIGSWLAWY QQKPGKAPKLLIYAADRLQSGVPSRFSGS GSGTDFTLTISSLQPEDFATYYCQQYHTY PLTFGGGTKVEIKAAGSDQEPKSSDKTHT CPPCSAPELLGGSSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSRDELTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
230	1198-A01	VH		EVQLVESGGGLVQPGGSLRLSCAASGFTF SDYDMSWVRQAPGKGLEWVGFIAQDGSYK YYVDSVKGRFTISRDNAKNSLYLQMNSLR AEDTAVYYCARSKLFRAGQFDYWGQGTLV TVSS

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231	1198-B10	VH		EVQLVESGGGLVQPGGSLRLSCAASGFNI SGSWIHWVRQAPGKGLEWVGYIYPDDGDT YYADSVKGRFTISADTSKNTAYLQMNSLR AEDTAVYYCAREGSHNLDKMDYWGQGTLV TVSS
232	1198-D03	VH		EVQLVESGGGLVQPGGSLRLSCAASGFNI NNYDIHWVRQAPGKGLEWVANIDPYNGAT YYADSVKGRFTISADTSKNTAYLQMNSLR AEDTAVYYCARVLWGFWAPFDYWGQGTLV TVSS
233	1198-D04	VH		EVQLVESGGGLVQPGGSLRLSCAASGFNI NNYDIHWVRQAPGKGLEWVANIDPYNGTT YYADSVKGRFTISADTSKNTAYLQMNSLR AEDTAVYYCARVPWGFWAPFDYWGQGTLV TVSS
234	1251-A02	VH		QVQLVESGGGVVQPGRSLRLSCAASGFAF SDHGMHWVRQAPDKGLEWVAVIWYDGSHK IYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGSLAGGAVYGTDVWGQG TTVTVSS
235	1251-A03	VH		QVQLVESGGGVVQPGRSLRLSCAASGFAF SDHGMHWVRQAPDKGLEWVAVIWYDGSHK IYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGSLAGGAVYGTDVWGQG TTVTVSS
236	1251-B08-g	VH		QVQLVESGGGVVQPGRSLRLSCAASGFNF SDYGMHWVRQAPGKGLEWVAVIWYDGSIS YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTVEHGAVYGTDVWGQG TTVTVSS
237	1251-A06-g	VH		QVQLVESGGGVVQPGRSLRLSCAASGFDF SSYGMHWVRQAPGKGLEWVAVIWDDGSDR YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTRVLGAIHGTDVWGQG TTVTVSS
238	1251-B08	VH		QVQLVESGGGVVQPGRSLRLSCAASGFNF SDYGMHWVRQAPDKGLEWVAVIWYDGSIS YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTVEHGAVYGTDVWGQG ATVTVSS
239	1251-A06	VH		QVQLVESGGGVVQPGRSLRLSCAASGFDF SSYGMHWVRQAPDKGLEWVAVIWDDGSDR YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTRVLGAIHGTDVWGQG TTVTVSS

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240	1251-B09	VH		QVQLVESGGGVVQPGRSLRLSCAASGFNF SDYGMHWVRQAPDKGLEWVAVTWYDGSRE YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTLVHGALYGNDVWGQG TTVTVSS
241	1251-B10	VH		QVQLVESGGGVVQPGRSLRLSCAASGFNF SSHGMHWVRQAPDKGLEWVAVIWHDGSDK YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTRVLGAVYGLDVWGQG TTVTVSS
242	1251-C03	VH		QVQLVESGGGVVQPGRSLRLSCAASGFNF SSYGMHWVRQAPDKGLEWVAVIWYDGSIK NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGALMRGEFSGHDVWGQG TTVTVSS
243	1251-D02	VH		QVQLVESGGGVVQPGRSLRLSCAASGFSF ASHGMHWVRQAPDKGLEWVAVIWDDGSDR YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTRVLGAIHGTDVWGQG TTVTVSS
244	1251-D06	VH		QVQLVESGGGVVQPGRSLRLSCAASGFSF GSYGMHWVRQAPDKGLEWVAVVWYDGSKT IYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTLVRGAVYGLDVWGQG TTVTVSS
245	1251-D09	VH		QVQLVESGGGVVQPGRSLRLSCAASGFSF SSYGMHWVRQAPDKGLEWVAVIWYDASIR KYAGSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTVERGAIYGTDVWGQG TTVTVSS
246	1251-E06	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF DSYGMHWVRQAPDKGLEWVAVIWYDGSNK VYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGMVGQGAMFGLDVWGQG TTVTVSS
247	1251-F06	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSFGMHWVRQAPDKGLEWVAVIWYDGSNE YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGSLVTRGVYGLDVWGQG TTVTVSS
248	1251-F07	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSHGMHWVRQAPDKGLEWVAVIWDDGSNE VYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTRIRGLRYGTDVWGQG TTVTVSS

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249	1251-G02	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSYGMHWVRQAPDKGLEWVAVIWHDGSKD YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGQLDHGAIYGLDVWGQG TTVTVSS
250	1445-A03	VH		EVQLVESGGGLVQPGGSLRLSCAASGFNI SGYYIHWVRQAPGKGLEWVAEISPTGGYT YYADSVKGRFTISADTSKNTAYLQMNSLR AEDTAVYYCAREHGLVYGQPMDYWGQGTL VTVSS
251	1445-B09	VH		EVQLVESGGGLVQPGGSLRLSCAASGFNI TGTGIHWVRQAPGKGLEWVGIITPYNGTT NYADSVKGRFTISADTSKNTAYLQMNSLR AEDTAVYYCARGGYGYYYPPFDYWGQGTL VTVSS
252	VH11-[19]	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSYGMHWVRQAPDKGLEWVAVIWYDGSNK YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGGTLVRGAMYGTDVWGQG TTVTVSS
253	VH5-[7]	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSYAMHWVRQAPGKGLEWVAVISYDGSNK YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCASGRYYGSGSYSSYFDYWGQ GTLVTVSS
254	VH6-[11]	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSYAMHWVRQAPGKGLEWVAVISYDGSIK YYADSVKGRFTISRDNSKNTLYLQMNSLR VEDTAVFYCARGREITSQNIVILLDYWGQ GTLVTVTS
255	VH8-[15]	VH		QVQLVESGGGVVQPGRSLRLSCAASGFTF SSYAMHWVRQAPGKGLEWVAVISYDGSNK YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCARGREITSQNIVILLDYWGQ GTLVTVSS
256	1337-A09-g	VL		DIQMTQSPSSVSASVGDRVTITCRASQGI GSWLAWYQQKPGKAPKLLIYAADRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYHTYPLTFGGGTKVEIK
257	1275-C10-g	VL		DIQMTQSPSSVSASVGDRVTITCRASQGV SSWLAWYQQKPGKAPKLLIYSARYLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNLYPLTFGGGTKVEIK

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258	1275-D01	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI GRWLAWYQQKPEKAPKSLIYGRSSLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNIYPLTFGGGTKVEIK
259	1275-D10	VL		DIQMTQSPSSLSASVGDRVTITCRASQGV FSWLAWYQQKPEKAPKSLIYNATQLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYYYYPLTFGGGTKVEIK
260	1275-G02	VL		DIQMTQSPSSLSASVGDRVTITCRASQGL GSFLAWYQQKPEKAPKSLIYLGNLLQIGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNAYPLTFGGGTKVEIK
261	1337-A04	VL		DIQMTQSPSSLSASVGDRVTITCRASQDI GRWVAWYQQKPEKAPKSLIYGASSLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNTYPLTFGGGTKVEIK
262	1337-A05	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI GRWVAWYQQKPEKAPKSLIYGADRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNSYPLTFGGGTKVEIK
263	1337-A06	VL		DIQMTQSPSSLSASVGDRVTITCRASQDI GSWVAWYQQKPEKAPKSLIYGADRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNSYPLTFGGGTKVEIK
264	1337-A07-g	VL		DIQMTQSPSSVSASVGDRVTITCRASQGI SSWVAWYQQKPGKAPKLLIYGASRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYHTYPLTFGGGTKVEIK
265	1337-A07	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI SSWVAWYQQKPEKAPKSLIYGASRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYHTYPLTFGGGTKVEIK
266	1337-A08	VL		DIQMTQSPSSLSASVGDRVTITCRASQDI GSWVAWYQQKPEKAPKSLIYASDSLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNSYPLTFGGGTKVEIK
267	1337-A09	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI GSWLAWYQQKPEKAPKSLIYAADRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYHTYPLTFGGGTKVEIK
268	1275-C10	VL		DIQMTQSPSSLSASVGDRVTITCRASQGV SSWLAWYQQKPEKAPKSLIYSARYLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNLYPLTFGGGTKVEIK

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269	1337-A10	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI SSWVAWYQQKPEKAPKSLIYGSSRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNTYPLTFGGGTKVEIK
270	1337-A10-g	VL		DIQMTQSPSSVSASVGDRVTITCRASQGI SSWVAWYQQKPGKAPKLLIYGSSRLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNTYPLTFGGGTKVEIK
271	VL5-[23]	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI SSWLAWFQQKPEKAPKSLIYAASSLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNSYPLTFGGGTKVEIK
272	VL6-[26]	VL		DIQMTQSPSSLSASVGDRVTITCRASQGI SSWLAWYQQKPEKAPKSLIYAASSLQSGV PSRFSGSGSGTDFTLTISSLQPEDFATYY CQQYNSYPLTFGGGTKVEIK
273	1193-B06	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF TGNWMSWVRQAPGKGLEWVGIIYGTSGAT YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKPSMSGSRGFDYWGQGTLV TVSS
274	1193-C08	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNNWMSWVRQAPGKGLEWVGIINGDDGYT YYADRVKGRFTIIRDNSKNTLYLQMNSLR AEDTAVYYCAKVALGRPRRFDYWGQGTLV TVSS
275	1193-E06b	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNTDMSWVRQAPGKGLEWVGIINGSGGAT NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKFENEWEVSMDYWGQGTLV TVSS
276	1193-H04b	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF TSSWMSWVRQAPGKGLEWVGIINGYNGIT YYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKPSAPGARRFDYWGQGTLV TVSS
277	1447-D11	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF NNTDMSWVRQAPGKGLEWVGVINGSGGSS NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYETEWEVSLDYWGQGTLV TVSS
278	1447-E08	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF NDTDMSWVRQAPGKGLEWVGMINGAGGAS FYADSVRGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKFENQWEVTFDYWGQGTLV TVSS

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279	1447-F11	VH		EVQLLESGGGLVQTGGSLRLSCAASGFTF DNTDMSWVRQAPGKGLEWVGIINGSGGVT NYADSVRGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYESEWEVSLDYWGQGTLV TVSS
280	1447-G01	VH		EVQLLESGGGLVQPGGSLRLSCAASGFTF NTSDMSWVRQAPGKGLEWVGIINGSGGAT NYADSVKGRFTISRDNSKNTLYLQMNSLR AEDTAVYYCAKYENEMEVSMDYWGQGTLV TVSS
281	1193-B06	VL		EIVLTQSPGTLSLSPGERATLSCRAGQSV SSSYLAWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQQHYTTPPTFGQGTKVEIK
282	1193-C08	VL		EIVLTQSPGTLSLTPGERATLSCRASQSV SSNYLAWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAMY YCQQHYTTPPTFGQGTKVEIK
283	1193-E06b	VL		EIVLTQSPGTLSLSPGERATLSCRASQSV SSSYLAWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQQHYTTPPTFGQGTKVEIK
284	1193-H04b	VL		EIVLTQSPGTLSLSPGEATLSCRASQSVS SSYLAWYQQRPGQAPRLLIYGASSRATGI PDRFSGSGSGTDFTLTISRLEPEDFAVYY CQQHYTTPPTFGQGTKVEIK
285	1447-D11	VL		EIVLTQSPGTLSLSPGERATLSCRASQSV SSSYLAWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQHNQPTPPTFGQGTKVEIK
286	1447-E08	VL		EIVLTQSPGTLSLSPGERATLSCRASQRV AGIDLSWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQQHNTTPPTFGQGTKVEIK
287	1447-F11	VL		EIVLTQSPGTLSLSPGERATLSCRASQSV YRSYLAWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQQHQTAPPTFGQGTKVEIK
288	1447-G01	VL		EIVLTQSPGTLSLSPGERATLSCRASQSV SSRELGWYQQKPGQAPRLLIYGASSRATG IPDRFSGSGSGTDFTLTISRLEPEDFAVY YCQQQCSWPPTFGQGTKVEIK

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289	IgG1 Fc from scFv-Fc			AAGSDQEPKSSDKTHTCPPCSAPELLGGS SVFLFPPKPKDTLMISRTPEVTCVVVDVS HEDPEVKFNWYVDGVEVHNAKTKPREEQY NSTYRVVSVLTVLHQDWLNGKEYKCKVSN KALPAPIEKTISKAKGQPREPQVYTLPPS RDELTKNQVSLTCLVKGFYPSDIAVEWES NGQPENNYKTTPPVLDSDGSFFLYSKLTV DKSRWQQGNVFSCSVMHEALHNHYTQKSL SLSPGK
290	Trastuzumab LC	LC		DIQMTQSPSSLSASVGDRVTITCRASQDV NTAVAWYQQKPGKAPKLLIYSASFLYSGV PSRFSGSRSGTDFTLTISSLQPEDFATYY CQQHYTTPPTFGQGTKVEIK
291	1251-A06-(wt)	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFD FSSYGMHWVRQAPDKGLEWVAVIWDDGSD RYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTRVLGAIHGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVEDYFPEPATVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
292	1251-A06-(g)	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFD FSSYGMHWVRQAPGKGLEWVAVIWDDGSD RYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTRVLGAIHGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK

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293	1251-B08-(wt)	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFN FSDYGMHWVRQAPDKGLEWVAVIWYDGSI SYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTVEHGAVYGTDVWGQ GATVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
294	1251-B08-(g)	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFN FSDYGMHWVRQAPGKGLEWVAVIWYDGSI SYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTVEHGAVYGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
295	1275-C10-(wt)	LC		MDIQMTQSPSSLSASVGDRVTITCRASQG VSSWLAWYQQKPEKAPKSLIYSARYLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYNLYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC

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296	1275-C10-(g)	LC		MDIQMTQSPSSVSASVGDRVTITCRASQG VSSWLAWYQQKPGKAPKLLIYSARYLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYNLYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
297	1337-A07-(wt)	LC		MDIQMTQSPSSLSASVGDRVTITCRASQG ISSWVAWYQQKPEKAPKSLIYGASRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYHTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
298	1337-A07-(g)	LC		MDIQMTQSPSSVSASVGDRVTITCRASQG ISSWVAWYQQKPGKAPKLLIYGASRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYHTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
299	1337-A09-(wt)	LC		MDIQMTQSPSSLSASVGDRVTITCRASQG IGSWLAWYQQKPEKAPKSLIYAADRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYHTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
300	1337-A09-(g)	LC		MDIQMTQSPSSVSASVGDRVTITCRASQG IGSWLAWYQQKPGKAPKLLIYAADRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYHTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC

SEQ ID NO	Molecule	Region	Scheme	Sequence
301	1337-A10-(wt)	LC		MDIQMTQSPSSLSASVGDRVTITCRASQG ISSWVAWYQQKPEKAPKSLIYGSSRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYNTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
302	1337-A10-(g)	LC		MDIQMTQSPSSVSASVGDRVTITCRASQG ISSWVAWYQQKPGKAPKLLIYGSSRLQSG VPSRFSGSGSGTDFTLTISSLQPEDFATY YCQQYNTYPLTFGGGTKVEIKRTVAAPSV FIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDSTY SLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC
303	C-term His Tag	Tag		GGSHHHHHH
304	HC Constant	HC Constant		ASTKGPSVFPLAPSSKSTSGGTAALGCLV KDYFPEPVTVSWNSGALTSGVHTFPAVLQ SSGLYSLSSVVTVPSSSLGTQTYICNVNH KPSNTKVDKKVEPKSCDKTHTCPPCPAPE LLGGPSVFLFPPKPKDTLMISRTPEVTCV VVDVSHEDPEVKFNWYVDGVEVHNAKTKP REEQYNSTYRVVSVLTVLHQDWLNGKEYK CKVSNKALPAPIEKTISKAKGQPREPQVY TLPPSREEMTKNQVSLTCLVKGFYPSDIA VEWESNGQPENNYKTTPPVLDSDGSFFLY SKLTVDKSRWQQGNVFSCSVMHEALHNHY TQKSLSLSPGK
305	LC Constant	LC Constant		RTVAAPSVFIFPPSDEQLKSGTASVVCLL NNFYPREAKVQWKVDNALQSGNSQESVTE QDSKDSTYSLSSTLTLSKADYEKHKVYAC EVTHQGLSSPVTKSFNRGEC

SEQ ID NO	Molecule	Region	Scheme	Sequence
306	1251-A03	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFA FSDHGMHWVRQAPDKGLEWVAVIWYDGSH KIYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGSLAGGAVYGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTR TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
307	1251-B09	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFN FSDYGMHWVRQAPDKGLEWVAVTWYDGSR EYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTLVHGALYGNDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK

SEQ ID NO	Molecule	Region	Scheme	Sequence
308	1251-B10	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFN FSSHGMHWVRQAPDKGLEWVAVIWHDGSD KYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTRVLGAVYGLDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSSTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK
309	Fc Constant	Fc Constant		AAGSDQEPKSSDKTHTCPPCSAPELLGGS SVFLFPPKPKDTLMISRTPEVTCVVVDVS HEDPEVKFNWYVDGVEVHNAKTKPREEQY NSTYRVVSVLTVLHQDWLNGKEYKCKVSN KALPAPIEKTISKAKGQPREPQVYTLPPS RDELTKNQVSLTCLVKGFYPSDIAVEWES NGQPENNYKTTPPVLDSDGSFFLYSKLTV DKSRWQQGNVFSCSVMHEALHNHYTQKSL SLSPGK
310	1251-A06 HC	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFD FSSYGMHWVRQAPDKGLEWVAVIWDDGSD RYYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTRVLGAIHGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK

SEQ ID NO	Molecule	Region	Scheme	Sequence
311	1251-F07 HC	HC		MQVQLVESGGGVVQPGRSLRLSCAASGFT FSSHGMHWVRQAPDKGLEWVAVIWDDGSN EVYADSVKGRFTISRDNSKNTLYLQMNSL RAEDTAVYYCARGGTRIRGLRYGTDVWGQ GTTVTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGV HTFPAVLQSSGLYSLSSVVTVPSSSLGTQ TYICNVNHKPSNTKVDKKVEPKSCDKTHT CPPCPAPELLGGPSVFLFPPKPKDTLMIS RTPEVTCVVVDVSHEDPEVKFNWYVDGVE VHNAKTKPREEQYNSTYRVVSVLTVLHQD WLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSREEMTKNQVSLTCLVK GFYPSDIAVEWESNGQPENNYKTTPPVLD SDGSFFLYSKLTVDKSRWQQGNVFSCSVM HEALHNHYTQKSLSLSPGK

Equivalents

[00402] 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.

WHAT IS CLAIMED IS:

1. An antibody conjugate comprising an antibody that specifically binds to CD74 linked site-specifically to at least one payload moiety, wherein the antibody comprises a non-natural amino acid at a site 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 wherein the antibody further comprises:

a. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 1 and 33; a CDR-H2 comprising at least one of SEQ ID NOs: 65 and 97; and a CDR-H3 comprising SEQ ID NO: 129;

b. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 2 and 34; a CDR-H2 comprising at least one of SEQ ID NOs: 66 and 98; and a CDR-H3 comprising SEQ ID NO: 130;

c. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 3 and 35; a CDR-H2 comprising at least one of SEQ ID NOs: 67 and 99; and a CDR-H3 comprising SEQ ID NO: 131;

d. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 4 and 36; a CDR-H2 comprising at least one of SEQ ID NOs: 68 and 100; and a CDR-H3 comprising SEQ ID NO: 132;

e. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 5 and 37; a CDR-H2 comprising at least one of SEQ ID NOs: 69 and 101; and a CDR-H3 comprising SEQ ID NO: 133;

f. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 6 and 38; a CDR-H2 comprising at least one of SEQ ID NOs: 70 and 102; and a CDR-H3 comprising SEQ ID NO: 134;

g. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 7 and 39; a CDR-H2 comprising at least one of SEQ ID NOs: 71 and 103; and a CDR-H3 comprising SEQ ID NO: 135;

h. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 8 and 40; a CDR-H2 comprising at least one of SEQ ID NOs: 72 and 104; and a CDR-H3 comprising SEQ ID NO: 136;

i. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 9 and 41; a CDR-H2 comprising at least one of SEQ ID NOs: 73 and 105; and a CDR-H3 comprising SEQ ID NO: 137;

j. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 10 and 42; a CDR-H2 comprising at least one of SEQ ID NOs: 74 and 106; and a CDR-H3 comprising SEQ ID NO: 138;

k. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 11 and 43; a CDR-H2 comprising at least one of SEQ ID NOs: 75 and 107; and a CDR-H3 comprising SEQ ID NO: 139;

l. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 12 and 44; a CDR-H2 comprising at least one of SEQ ID NOs: 76 and 108; and a CDR-H3 comprising SEQ ID NO: 140;

m. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 13 and 45; a CDR-H2 comprising at least one of SEQ ID NOs: 77 and 109; and a CDR-H3 comprising SEQ ID NO: 141;

n. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 14 and 46; a CDR-H2 comprising at least one of SEQ ID NOs: 78 and 110; and a CDR-H3 comprising SEQ ID NO: 142;

o. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 15 and 47; a CDR-H2 comprising at least one of SEQ ID NOs: 79 and 111; and a CDR-H3 comprising SEQ ID NO: 143;

p. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 16 and 48; a CDR-H2 comprising at least one of SEQ ID NOs: 80 and 112; and a CDR-H3 comprising SEQ ID NO: 144;

q. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 17 and 49; a CDR-H2 comprising at least one of SEQ ID NOs: 81 and 113; and a CDR-H3 comprising SEQ ID NO: 145;

r. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 18 and 50; a CDR-H2 comprising at least one of SEQ ID NOs: 82 and 114; and a CDR-H3 comprising SEQ ID NO: 146;

s. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 19 and 51; a CDR-H2 comprising at least one of SEQ ID NOs: 83 and 115; and a CDR-H3 comprising SEQ ID NO: 147;

t. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 20 and 52; a CDR-H2 comprising at least one of SEQ ID NOs: 84 and 116; and a CDR-H3 comprising SEQ ID NO: 148;

u. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 21 and 53; a CDR-H2 comprising at least one of SEQ ID NOs: 85 and 117; and a CDR-H3 comprising SEQ ID NO: 149;

v. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 22 and 54; a CDR-H2 comprising at least one of SEQ ID NOs: 86 and 118; and a CDR-H3 comprising SEQ ID NO: 150;

w. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 23 and 55; a CDR-H2 comprising at least one of SEQ ID NOs: 87 and 119; and a CDR-H3 comprising SEQ ID NO: 151;

x. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 24 and 56; a CDR-H2 comprising at least one of SEQ ID NOs: 88 and 120; and a CDR-H3 comprising SEQ ID NO: 152;

y. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 25 and 57; a CDR-H2 comprising at least one of SEQ ID NOs: 89 and 121; and a CDR-H3 comprising SEQ ID NO: 153;

z. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 26 and 58; a CDR-H2 comprising at least one of SEQ ID NOs: 90 and 122; and a CDR-H3 comprising SEQ ID NO: 154;

aa. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 27 and 59; a CDR-H2 comprising at least one of SEQ ID NOs: 91 and 123; and a CDR-H3 comprising SEQ ID NO: 155; or

bb. a V_H comprising: a CDR-H1 comprising at least one of SEQ ID NOs: 28 and 60; a CDR-H2 comprising at least one of SEQ ID NOs: 92 and 124; and a CDR-H3 comprising SEQ ID NO: 156.

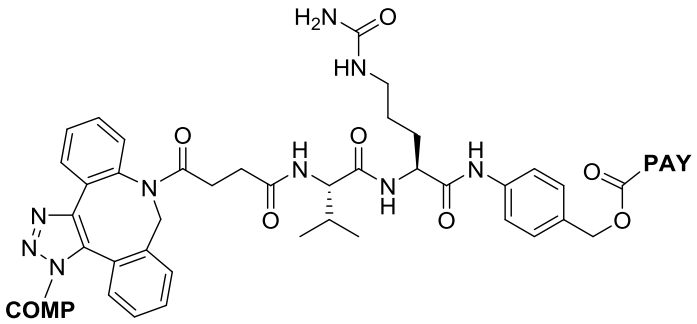
2. The antibody conjugate of claim 1 or 2, wherein the non-natural amino acid is selected from the group consisting of p-acetyl-L-phenylalanine, O-methyl-L-tyrosine, an -3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, a tri-O-acetyl-GlcNAcβ-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-azido-L-phenylalanine, p-azidomethyl-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.

3. The antibody conjugate of claim 2, wherein the antibody is linked to the payload moiety via a linker that is hydrolytically stable.

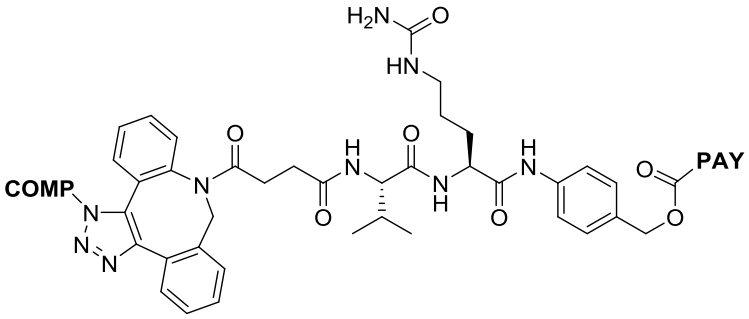
4. The antibody conjugate of claim 2, wherein the antibody is linked to the payload moiety via a linker that is cleavable.

5. The antibody conjugate of any one of claims 2 to 4 wherein the non-natural amino acid residue is according to compound 30 or compound 56.

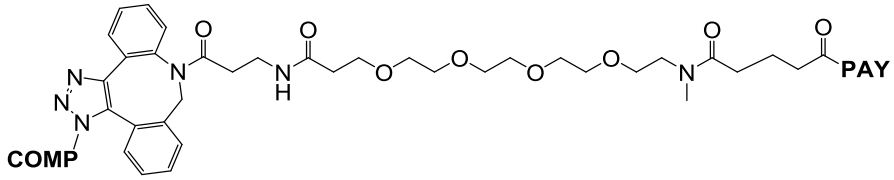
6. The antibody conjugate of any of the preceding claims according to any of formulas 101a-104b:



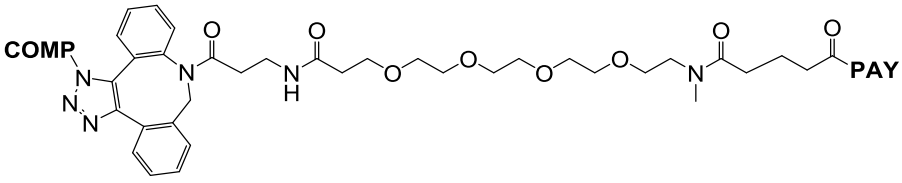
(101a)



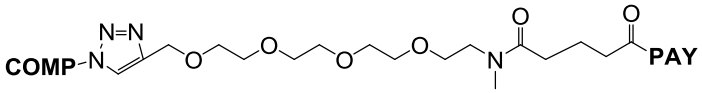
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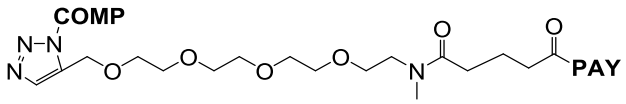
(102a)



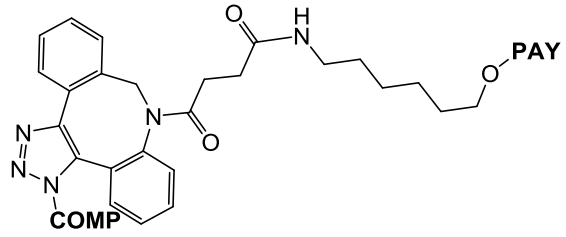
(102b)



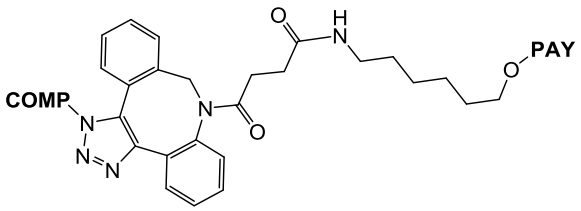
(103a)



(103b)



(104a)



(104b)

wherein **COMP** is an amino acid residue of the antibody bearing the indicated bond and **PAY** is a payload moiety.

7. The antibody conjugate of any of the preceding claims wherein the payload moiety is selected from the group consisting of maytansines, hemiasterlins, amanitins, and auristatins.

8. The antibody conjugate of any of the preceding claims wherein the payload moiety is selected from the group consisting of DM1, hemiasterlin, amanitin, MMAF, and MMAE.

9. The antibody conjugate of any of the preceding claims, wherein the antibody comprises:

a. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 161; a CDR-L2 comprising SEQ ID NO: 181; and a CDR-L3 comprising SEQ ID NO: 201;

b. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 162; a CDR-L2 comprising SEQ ID NO: 182; and a CDR-L3 comprising SEQ ID NO: 202;

c. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 163; a CDR-L2 comprising SEQ ID NO: 183; and a CDR-L3 comprising SEQ ID NO: 203;

d. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 164; a CDR-L2 comprising SEQ ID NO: 184; and a CDR-L3 comprising SEQ ID NO: 204;

e. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 165; a CDR-L2 comprising SEQ ID NO: 185; and a CDR-L3 comprising SEQ ID NO: 205;

f. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 166; a CDR-L2 comprising SEQ ID NO: 186; and a CDR-L3 comprising SEQ ID NO: 206;

g. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 167; a CDR-L2 comprising SEQ ID NO: 187; and a CDR-L3 comprising SEQ ID NO: 207;

h. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 168; a CDR-L2 comprising SEQ ID NO: 188; and a CDR-L3 comprising SEQ ID NO: 208;

i. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 169; a CDR-L2 comprising SEQ ID NO: 189; and a CDR-L3 comprising SEQ ID NO: 209;

j. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 170; a CDR-L2 comprising SEQ ID NO: 190; and a CDR-L3 comprising SEQ ID NO: 210;

k. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 171; a CDR-L2 comprising SEQ ID NO: 191; and a CDR-L3 comprising SEQ ID NO: 211;

l. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 172; a CDR-L2 comprising SEQ ID NO: 192; and a CDR-L3 comprising SEQ ID NO: 212;

m. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 173; a CDR-L2 comprising SEQ ID NO: 193; and a CDR-L3 comprising SEQ ID NO: 213;

n. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 174; a CDR-L2 comprising SEQ ID NO: 194; and a CDR-L3 comprising SEQ ID NO: 214;

o. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 175; a CDR-L2 comprising SEQ ID NO: 195; and a CDR-L3 comprising SEQ ID NO: 215;

p. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 176; a CDR-L2 comprising SEQ ID NO: 196; and a CDR-L3 comprising SEQ ID NO: 216;

q. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 177; a CDR-L2 comprising SEQ ID NO: 197; and a CDR-L3 comprising SEQ ID NO: 217;

r. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 178; a CDR-L2 comprising SEQ ID NO: 198; and a CDR-L3 comprising SEQ ID NO: 218; or

s. a V_L comprising: a CDR-L1 comprising SEQ ID NO: 179; a CDR-L2 comprising SEQ ID NO: 199; and a CDR-L3 comprising SEQ ID NO: 219.

10. The antibody conjugate of any of the preceding claims, wherein the antibody comprises:

a. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 256, or the variant thereof;

b. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 257, or the variant thereof;

c. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 265, or the variant thereof;

d. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 264, or the variant thereof;

e. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 267, or the variant thereof;

f. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 268, or the variant thereof;

g. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 269, or the variant thereof;

h. the V_H region is SEQ ID NO: 236, or the variant thereof, and the V_L region is SEQ ID NO: 270, or the variant thereof;

i. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 256, or the variant thereof;

j. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 257, or the variant thereof;

k. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 265, or the variant thereof;

l. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 264, or the variant thereof;

m. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 267, or the variant thereof;

n. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 268, or the variant thereof;

o. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 269, or the variant thereof;

p. the V_H region is SEQ ID NO: 237, or the variant thereof, and the V_L region is SEQ ID NO: 270, or the variant thereof;

q. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 256, or the variant thereof;

r. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 257, or the variant thereof;

s. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 265, or the variant thereof;

t. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 264, or the variant thereof;

u. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 267, or the variant thereof;

v. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 268, or the variant thereof;

w. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 269, or the variant thereof;

x. the V_H region is SEQ ID NO: 238, or the variant thereof, and the V_L region is SEQ ID NO: 270, or the variant thereof;

y. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 256, or the variant thereof;

z. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 257, or the variant thereof;

aa. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 265, or the variant thereof;

bb. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 264, or the variant thereof;

cc. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 267, or the variant thereof;

dd. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 268, or the variant thereof;

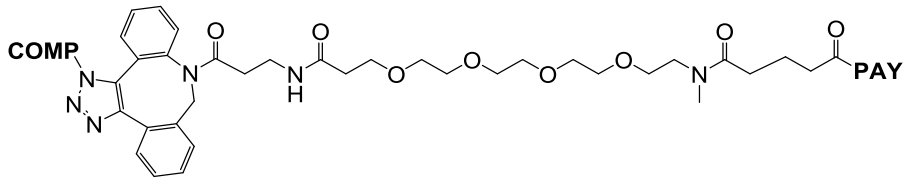
ee. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 269, or the variant thereof; or

ff. the V_H region is SEQ ID NO: 239, or the variant thereof, and the V_L region is SEQ ID NO: 270, or the variant thereof.

11. The antibody conjugate of claim 10, wherein the antibody comprises a non-natural amino acid at site HC-F404 according to the EU numbering scheme of Kabat.

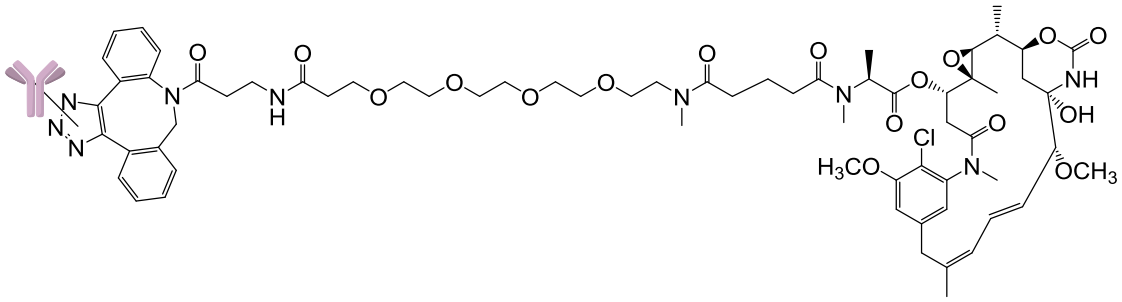
12. The antibody conjugate of claim 11, wherein the non-natural amino acid is para-azidomethylphenylalanine or p-azidomethyl-L-phenylalanine.

13. The antibody conjugate of claim 12, wherein the antibody conjugate has the formula of (102b):



wherein **COMP** is the non-natural amino acid residue of the antibody bearing the indicated bond and **PAY** is DM1.

14. The antibody conjugate of claim 13, wherein the antibody conjugate has the structure of **Conjugate A**:



and the antibody comprises a V_H region comprising SEQ ID NO: 236 and a V_L region comprising SEQ ID NO: 256.

15. The antibody conjugate of any of the preceding claims, further comprising at least one constant region domain.

16. The antibody conjugate of claim 15, wherein the constant region comprises a sequence selected from SEQ ID NOs: 304-305.

17. The antibody conjugate of any of the preceding claims, wherein the antibody is a monoclonal antibody.

18. The antibody of any of the preceding claims, wherein the antibody is an IgA, an IgD, an IgE, an IgG, or an IgM.

19. The antibody conjugate of any of the preceding claims, wherein the antibody is humanized or human.

20. The antibody conjugate of any of the preceding claims, wherein the antibody is aglycosylated.

21. The antibody conjugate of any of the preceding claims, wherein the antibody is an antibody fragment.

22. The antibody conjugate of claim 21, 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.

23. The antibody conjugate of claim 22, wherein the antibody is an scFv fragment.

24. The antibody conjugate of claim 23, wherein the scFv fragment comprises a sequence selected from SEQ ID NOs: 221-228.

25. The antibody conjugate of claim 22, wherein the antibody is an scFv-Fc fragment.

26. The antibody conjugate of claim 25, wherein the scFv-Fc fragment comprises SEQ ID NO: 229.

27. The antibody conjugate of any of the preceding claims, wherein the antibody has a k_a of at least about 10^5 $M^{-1}\times sec^{-1}$ at a temperature of 25°C.

28. The antibody conjugate of any of the preceding claims, wherein the antibody has a k_d of 10^{-3} sec^{-1} or less at a temperature of 25°C.

29. The antibody conjugate of any of the preceding claims, wherein the antibody has a K_D of 10^{-9} M or less at a temperature of 25°C.

30. The antibody conjugate of any of the preceding claims, wherein the antibody is internalized after binding to CD74 on the surface of a cell.

31. The antibody conjugate of any of the preceding claims, wherein the Tm2 of the antibody is at least 75°C, 75.5°C, 76°C, 76.5°C, 77°C, 77.5°C, 78°C, 78.5°C, or 79°C.

32. The antibody conjugate of any of the preceding claims, wherein the Tm1 of the antibody is less than 61°C or less than 60°C.

33. A kit comprising an antibody conjugate of any of the preceding claims, and instructions for use of the antibody conjugate.

34. The kit of claim 33, wherein the antibody conjugate is lyophilized.

35. The kit of claim 34, further comprising a fluid for reconstitution of the lyophilized antibody conjugate.

36. A pharmaceutical composition comprising the antibody conjugate of any of claims 1 to 32 and a pharmaceutically acceptable carrier.

37. An antibody conjugate of any one of claims 1 to 32, or a pharmaceutical composition of claim 36 for use in treating or preventing a disease or condition in a subject.

38. An antibody conjugate of any one of claims 1 to 32, or a pharmaceutical composition of claim 36 for use in diagnosing a disease or condition in a subject.

39. The antibody conjugate or pharmaceutical composition of any of claims 37 or 38, wherein the disease or condition is selected from a cancer, an autoimmune disease, an inflammatory disease, or an infection.

40. The antibody conjugate or pharmaceutical composition of claim 39, wherein the cancer is selected from multiple myeloma or pancreatic cancer.

41. The antibody conjugate or pharmaceutical composition of claim 39, wherein the disease or condition is B-cell lymphoma.

42. The antibody conjugate or pharmaceutical composition of claim 39, wherein the disease or condition is non-Hodgkin’s lymphoma.

43. The antibody conjugate or pharmaceutical composition of claim 39, wherein the disease or condition is leukemia.