(19) DANMARK

(10) DK/EP 3355913 T3

(12) Oversættelse af europæisk patentskrift

Patent- og Varemærkestyrelsen

(51) Int.Cl.: *A 61 K 39/00 (2006.01)* *C 07 K 16/28 (2006.01)* *C 07 K 16/46 (2006.01)*

(45) Oversættelsen bekendtgjort den: **2024-12-02**

(80) Dato for Den Europæiske Patentmyndigheds bekendtgørelse om meddelelse af patentet: **2024-10-30**

(86) Europæisk ansøgning nr.: **16784320.0**

(86) Europæisk indleveringsdag: **2016-09-30**

(87) Den europæiske ansøgnings publiceringsdag: **2018-08-08**

(86) International ansøgning nr.: **US2016055041**

(87) Internationalt publikationsnr.: **WO2017059380**

(30) Prioritet: **2015-09-30 US 201562235518 P**

(84) Designerede stater: **AL AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HR HU IE IS IT LI LT LU LV MC MK MT NL NO PL PT RO RS SE SI SK SM TR**

(73) Patenthaver: **IGM Biosciences, Inc., 325 East Middlefield Road, Mountain View, CA 94043, USA**

(72) Opfinder: **KEYT, Bruce, 1180 Lakeview Drive, Hillsborough, California 94010, USA**
PRESTA, Leonard George, 1900 Gough Street Apartment 206, San Francisco, California 94109, USA
BALIGA, Ramesh, 2237 Hopkins Avenue, Redwood City, CA 94062, USA

(74) Fuldmægtig i Danmark: **Budde Schou A/S, Dronningens Tværgade 30, 1302 København K, Danmark**

(54) Benævnelse: **BINDINGSMOLEKYLER MED MODIFICERET J-KÆDE**

(56) Fremdragne publikationer:
WO-A1-2015/153912
WO-A1-98/30591
US-A1- 2008 145 420
CHINTALACHARUVU K R ET AL: "Hybrid IgA2/IgG1 antibodies with tailor-made effector functions", CLINICAL IMMUNOLOGY, ACADEMIC PRESS, US, vol. 101, no. 1, 1 October 2001 (2001-10-01), pages 21 - 31, XP002999715, ISSN: 1521-6616, DOI: 10.1006/CLIM.2001.5083
SASKIA MEYER ET AL: "Improved in vivo anti-tumor effects of IgA-Her2 antibodies through half-life extension and serum exposure enhancement by FcRn targeting", MABS, vol. 8, no. 1, 14 October 2015 (2015-10-14), US, pages 87 - 98, XP055335017, ISSN: 1942-0862, DOI: 10.1080/19420862.2015.1106658
EL-RASHDYM REDWANA ET AL: "RecombinanthumanJ-chain:Fixtheprotein aggregationsandyieldmaximize", 1 January 2006 (2006-01-01), pages 1595 - 102, XP055333807, Retrieved from the Internet <URL:https://www.researchgate.net/profile/Elrashdy_Redwan/publication/282979117_Recombinant_human_J-chain_Fix_the_protein_aggregations_and_yield_maximize/links/57c9c8b708ae89cd1e81ece3.pdf>
"Current Protocols in Protein Science", 15 May 2001, JOHN WILEY & SONS, INC., US, ISSN: 1934-3655, article JOHANNES U. AMMANN ET AL: "Development and Use of IgM/J-Chain Fusion Proteins for Characterization of Immunoglobulin Superfamily Ligand-Receptor Interactions", pages: 19.24.1 - 19.24.11, XP055333814, DOI: 10.1002/0471140864.ps1924s75

Fortsættes ...

JANTINE E. BAKEMA ET AL: "Immunoglobulin A", MABS, vol. 3, no. 4, 1 July 2011 (2011-07-01), pages 352 - 361, XP055185206, ISSN: 1942-0862, DOI: 10.4161/mabs.3.4.16092

Description

FIELD OF THE INVENTION

[0001] The present invention concerns binding molecules that comprise an IgM, IgA, IgG/IgM or IgG/IgA antibody comprising a modified J-chain.

BACKGROUND OF THE INVENTION

[0002] J-chain is an acidic 15-kDa polypeptide, which is associated with pentameric IgM and dimeric IgA via disulfide bonds involving the penultimate cysteine residue in the 18-amino acid secretory tail-piece (tp) at the C-terminus of the IgM μ or IgA α heavy chain. The three disulfide bridges are formed between Cys 12 and 100, Cys 71 and 91, and Cys 108 and 133, respectively. *See, e.g.,* Frutiger et al. 1992, Biochemistry 31, 12643-12647. Structural requirements for incorporation of the J-chain into human IgM and IgA and for polymeric immunoglobulin assembly and association with the J-chain are reported by Sorensen et al. 2000, Int. Immunol. 12(1): 19-27 and Yoo et al. 1999, J. Biol. Chem. 274(47):33771-33777, respectively. Recombinant production of soluble J-chain in E coli is reported by Redwan et al. 2006, Human Antibodies 15:95-102. Recombinant J-chain constructs are also described in WO 98/30591 A1 (Epicyte Pharmaceutical Inc).

[0003] Methods for making hybrid IgA/IgG and IgM/IgG antibodies are known in the art. Thus, recombinant production of hybrid IgA2/IgG1 antibodies is reported in Chintalacharuvu et al. 2001, Clin Immunol 101(1):21-31. It has been reported that addition of αtp or μtp at the end of IgGγ heavy chain facilitates polymerization and enhances effector function such as complement activation (Smith et al., J Immunol 1995, 154:2226-2236). The IgA/IgG hybrid antibodies possess properties of both IgA and IgG. Methods for recombinant production of IgM antibodies are also known in the art. *E.g.,* Tchoudakova A, et al., High level expression of functional human IgMs in human PER.C6 cells. mAbs. 2009;1(2):163-171.

[0004] Despite the advances made in the design of antibodies, there remains a need for modified antibodies with improved properties, such as improved affinity, specificity and/or avidity, as well as the ability to bind to multiple binding targets.

[0005] As the field has progressed, antibody function has been enhanced through creative means of protein engineering, such as to provide higher affinity, longer half-life, and/or better tissue distribution, as well as combination of small and large molecule technologies for increased focus of cell destruction via toxic payload delivery (e.g., antibody-drug conjugates). Another approach to improving antibody function takes advantage of the bivalent binding of the immunoglobulin G (IgG) structure which allows one IgG molecule to bind two antigens. Indeed, in certain applications, there exists good potential for asymmetric antibodies to exert useful functions by simultaneously binding two different target antigens. To address this need, a variety of constructs have been produced to yield a single molecule that can bind two different antigens, allowing for functions never before seen in nature. An example of this bi-specific approach is "blinatumomab" (MT103 or AMG103) which binds the CD3 and CD19 receptors, on T- and B-cells, respectively. This tethering of a cytotoxic T-cell to a cancerous B-cell, allows for effective treatment of B-cell leukemia.

[0006] The blockade of immune checkpoints has emerged as a promising area for the advancement of cancer treatment. Immune checkpoints refer to inhibitory signaling pathways that are encoded into the immune system, and which play a vital role in maintaining self-tolerance, as well as modulating the duration and amplitude of immune responses. *See, e.g.,* Pardoll, Drew M. "The blockade of immune checkpoints in cancer immunotherapy." Nature Reviews Cancer 12.4 (2012): 252-264; Postow, Michael A. et al., "Immune Checkpoint Blockade in Cancer Therapy," J Clin Oncol. 2015 Jun 10;33(17): 1974-82. doi: 10.1200/JCO.2014.59.4358.

[0007] Despite positive proof of concept results in preclinical models, investigators have reported that monoclonal IgG blocking antibodies directed against T-cell inhibitory signaling pathway components (for example, ipilimumab (Bristol-Myers Squibb) and tremelimumab (MedImmune/AstraZenica), both directed against CTLA4) have only achieved minimal efficacy results in a clinical setting. *E.g., Postow* et al., pp. 1-2. In addition, treatments involving monoclonal IgG antibodies have resulted in immune-related adverse events, such as dermatologic, GI, hepatic, endocrine and other inflammatory events. *E.g., Id. at* p.4. As such, the use of monoclonal IgG antibodies in immune checkpoint blockade may be limited by the therapeutic index of such molecules, in that the dose of a monoclonal IgG antibody required to elicit the desired therapeutic effect also causes immune-related adverse events.

[0008] Accordingly, there is a need for binding molecules with increased avidity that will provide increased potency so that lower dosage levels can be used, thereby preventing the occurrence of immune-related adverse events, while still achieving effective blockade of T-cell inhibitory signaling pathways.

[0009] The pharmacokinetics and pharmacodynamics of monoclonal antibodies are complex, and depend on both the structure of the monoclonal antibody, as well as the physiological system that it targets. Moreover, different antibody classes are typically processed within a subject via different cellular and physiological systems. For example, secretion into the bile is an important pathway of elimination for IgA antibodies, whereas this route is not a significant contributor

to the elimination of IgG antibodies. Rather, the majority of IgG elimination occurs via intracellular catabolism, following fluid-phase or receptor-mediated endocytosis. *E.g.*, Wang et al., Nature 84:5 (2008). Furthermore, full-length IgG antibodies have been shown to be primarily distributed within the blood stream, while smaller IgG antibody fragments appear to distribute within the extra-vascular space to a greater extent. *E.g.,* Tabrizi et al., AAPSJ. 2010 Mar; 12(1): 33-43. The blood brain barrier generally prevents immunoglobulin molecules from entering the central nervous system via the circulation. *E.g.,* Yu et al., Science Translational Medicine 16:261 (2014). Furthermore, immunoglobulins that are directly injected into an extra-vascular space, such as the eyeball, typically only remain in the space on the order of hours. *See., e.g.,* Mordenti, J. et al., Toxicological Sciences 52, 101-106 (1999); Mordenti, J. et al., Toxicological Sciences 27(5), 536-544 (1999). As such, control and manipulation of factors that influence the absorption, distribution, metabolism and/or excretion (ADME) characteristics of monoclonal antibodies is an important consideration when designing a therapeutic antibody composition.

[0010] Accordingly, there is a need for binding molecules whose ADME characteristics can be controlled and modulated to achieve a desired therapeutic effect.

SUMMARY OF THE INVENTION

[0011] The present invention is based, at least in part, on the recognition that the J-chain of an IgM or IgA antibody can be modified by introducing one or more ADME-modulating moieties into a native J-chain sequence, and the modified J-chain can be introduced into IgM, IgA, IgG/IgM or IgG/IgA antibodies without compromising the functionality of the recipient antibody or the ADME-modulating moiety. This allows the IgM, IgA, IgG/IgM or IgG/IgA antibody to achieve improved properties, such as an increased concentration and/or an increased half-life in a subject.

[0012] The invention is further based on the recognition that due to their multivalent nature, IgM, IgA, IgG/IgM or IgG/IgA antibodies can provide increased avidity between the antibody and a target antigen, thereby facilitating binding of antigens with low level expression and/or low binding affinity. Furthermore, the optional multi-specific nature of the IgM, IgA, IgG/IgM or IgG/IgA portion of the subject binding molecules allows binding between specific numbers and/or specific types of binding targets, thereby facilitating binding between specific combinations of antigen targets. The modified J-chain portion of the subject binding molecules comprises an ADME-modulating moiety, which facilitates an increased concentration and/or an increased half-life in a target tissue.

[0013] An aspect of the invention includes a binding molecule comprising an IgM, IgA, IgG/IgM or IgG/IgA antibody with a modified J-chain, wherein the IgG/IgM or IgG/IgA antibody contains an IgM or IgA tail-piece at the IgG heavy chain, wherein the modified J-chain comprises an ADME-modulating moiety that reduces clearance of the antibody from a subject's circulation, wherein the ADME-modulating moiety is located at the C-terminus or the N-terminus of the modified J chain, and wherein the ADME-modulating moiety comprises an albumin protein, a fragment of an albumin protein, an albumin-binding peptide, an albumin-binding antibody fragment, an FcRn-binding peptide, or an FcRn-binding antibody fragment.

[0014] In some embodiments, the ADME-modulating moiety comprises human serum albumin.

[0015] In some embodiments, the IgM, IgA, IgG/IgM or IgG/IgA antibody binds to a hematologic cancer target which is CD20.

[0016] An aspect of the invention includes a binding molecule comprising an IgM, IgA, IgG/IgM or IgG/IgA antibody with a modified J-chain, wherein the IgG/IgM or IgG/IgA antibody contains an IgM or IgA tail-piece at the IgG heavy chain, wherein the modified J-chain comprises an ADME-modulating moiety that increases a concentration of the binding molecule in a central nervous system tissue of a subject, wherein the ADME-modulating moiety is located at the C-terminus or the N-terminus of the modified J chain, and wherein the ADME-modulating moiety comprises a transferrin protein, a leptin protein, a transferrin receptor-binding antibody fragment, a transferrin-binding antibody fragment, an insulin receptor-binding antibody fragment, an IGF-1 receptor-binding antibody fragment, or a leptin receptor-binding antibody fragment.

[0017] In some embodiments, the ADME-modulating moiety comprises a transferrin protein or a leptin protein.

[0018] An aspect of the invention includes a binding molecule comprising an IgM, IgA, IgG/IgM or IgG/IgA antibody with a modified J-chain, wherein the IgG/IgM or IgG/IgA antibody contains an IgM or IgA tail-piece at the IgG heavy chain, wherein the modified J-chain comprises an ADME-modulating moiety that increases retention of the binding molecule in an extra-vascular space of a subject, wherein the ADME-modulating moiety is located at the C-terminus or the N-terminus of the modified J chain, and wherein the ADME-modulating moiety comprises a hyaluronic acid binding protein (HABP), a hyaluronic acid-binding antibody fragment, a TSG-6 protein, or a TSG-6-binding antibody fragment.

[0019] In some embodiments, the IgM, IgA, IgG/IgM or IgG/IgA antibody binds to beta-secretase 1 (BACE).

[0020] In some embodiments, the modified J-chain comprises the native human J-chain sequence of SEQ ID NO: 1. In some embodiments, the ADME-modulating moiety is introduced into the native human J-chain sequence of SEQ ID NO: 1 by direct or indirect fusion, and indirect fusion is through a peptide linker. In some embodiments, the ADME-modulating moiety is introduced into the native human J-chain sequence of SEQ ID NO: 1 by chemical or chemo-

enzymatic derivatization. In some embodiments, the ADME-modulating moiety is introduced into the native human J-chain by a cleavable or non-cleavable linker, wherein the cleavable linker is a chemically labile linker or an enzyme-labile linker. In some embodiments, the linker is selected from the group consisting of N-succinimidyl-3-(2-pyridyldithio) propionate (SPDP), succinimidyl-4-(N-maleimidomethyl) cyclohexane-1-carboxylate (SMCC), N-succinimidyl-4-(2-pyridylthio) pentanoate (SPP), iminothiolane (IT), bifunctional derivatives of imidoesters, active esters, aldehydes, bis-azido compounds, bis-diazonium derivatives, diisocyanates, and bis-active fluorine compounds.

[0021] An aspect of the invention includes the binding molecule according to any one of the other aspects of the invention for use in treating cancer. In some embodiments, the cancer is a hematologic cancer, an epithelial cancer or a central nervous system cancer. Any reference to a method of treatment practised on the human or animal body is to be interpreted as substances and compositions for use in such treatments.

BRIEF DESCRIPTION OF THE DRAWINGS

[0022]

FIG. 1 illustrates the structure of an IgM pentamer, comprising a J-chain, wherein chains A and B are identical in native IgM.

FIG. 2 shows the schematic structures of IgA, dimeric IgA with J-chain, and dimeric J-chain integrated IgA with secretory IgA (sIgA).

FIG. 3 shows the amino acid sequence of mature human J-chain (SEQ ID NO: 1).

FIG. 4A illustrates two different orientations of J-chain constructs comprising a modified J-chain with a moiety that binds to CD3. The top illustration is an example of a modified J-chain that is in the J-linker-V orientation, with a binding moiety (e.g., an anti-CD3 scFv antibody fragment) positioned at the C-terminus of the modified J-chain. The bottom illustration is an example of a modified J-chain that is in the V-linker-J orientation, with a binding moiety (e.g., an anti-CD3 scFv antibody fragment) positioned at the N-terminus of the modified J-chain.

FIG. 4B illustrates two different orientations of J-chain constructs comprising a modified J-chain with an HSA-containing moiety. The top illustration is an example of a modified J-chain that is in the J-linker-ADME orientation, with an ADME modulating moiety (e.g., a human serum albumin (HSA) polypeptide) positioned at the C-terminus of the modified J-chain. The bottom illustration is an example of a modified J-chain that is in the ADME-linker-J orientation, with an ADME modulating moiety (e.g., a human serum albumin (HSA) polypeptide) positioned at the N-terminus of the modified J-chain.

FIG. 5 is a schematic illustration of an asymmetric IgM pentamer with binding specificity for a target antigen, comprising an ADME-modulating moiety fused to the J-chain at one end, and a CD3 binding moiety at the opposite end of the J-chain.

FIG. 6 shows SDS PAGE analysis of anti-CD20 IgM antibodies with or without various anti-CD3 binding moieties on the J-chain in either orientation. J-chain containing IgM pentamers are easily distinguished from the hexameric IgM without J-chain.

FIG. 7 is a graph showing cell viability as a function of antibody concentration for various antibody constructs in a complement dependent cytotoxicity assay in the presence of IgG, IgM or IgM's carrying various J-chains. A table is provided with the EC50 values for each construct.

FIG. 8 is a graph showing results of a T-cell activation assay comparing the ability of an anti-CD20 IgM with a CD3 binding moiety on the J-chain to activate T-cells, as compared to anti-CD20 IgM antibodies without a CD3 binding moiety on the J-chain, as well as anti-CD20 IgG antibodies.

FIG. 9, Panel A is a graph showing IgM concentration in mice in the absence of half-life extension for CDIM binding IgM 55.5. Panel B is a table providing PK parameters.

FIG. 10 is a graph showing results of a multimer specific ELISA for anti-CD20 IgM antibodies demonstrating the vastly tighter binding of IgM.

FIG. 11, Panel A shows an illustration of a temporal biodistribution model. Panel B shows data for the biodistribution of IGM-55.5 in vivo using conjugated far infra-red dye Vivo Tag 680 (Perkin Elmer).

FIG. 12 Panel A is a schematic illustration depicting site specific labeling of glycans on IgG using chemoenzymatic approach. Panel B shows the position of glycans on IgM heavy chain and J-chain. Panel C shows the non-reduced and reduced gels for the labled products after using chemo-enzymatic labeling.

FIG. 13 lists IgM, IgA, IgG/IgM or IgG/IgA antibody targets and ADME-modulating moieties that can be placed on the J-chain. Any of the antibody targets listed in the left column can be combined with any of the ADME-modulating moieties on a J-chain listed in the right column.

FIG. 14 is an illustration of the structure of Tn antigen.

FIG. 15 is an illustration of the structure of hyaluronic acid.

FIG. 16, Panel A, is a graph showing antibody concentration as a function of time in a BALB/c mouse PK experiment

for a model IgG (Rituximab), serum derived polyclonal IgM from humans, and an engineered CHO cell derived IgM (55.5). Panel B is a table showing alpha and beta half-life, and AUC for these three different antibodies.

FIG. 17, Panel A, is a graph showing antibody concentration as a function of time in a PK experiment in BALB/c mice testing the effect of J-chain incorporation in IgM. Panel B is a table showing alpha and beta half-life, and AUC for three different IgM antibodies with wild type (wt) or J-chain fused with an scFv configured to bind T-cells.

FIG. 18 is a graph showing serum concentration as a function of time for three different model antibodies: Rituximab(IgG); an anti-CD20 IgM with a domain configured to bind T-cells fused to the N-terminus of the J-chain; and an anti-CD20 IgM with an albumin binding domain (ABD) fused to the N-terminus of the J-chain with a 15-amino acid linker (A15J).

FIG. 19 is an image of a reducing PAGE gel and a Western blot analysis of the antibodies listed in the table. Incorporation of the J-chain with or without fused human serum albumin in either orientation with respect to J-chain is visualized using western blotting with an anti-J chain antibody.

FIG. 20, is a graph showing CDC activity as a function of concentration for four IgM antibodies demonstrating that incorporating a moiety as large as 65 KDa HSA does not disrupt the CDC activity of IgM.

FIG. 21, Panel A, is a graph showing concentration as a function of time in a mouse pharmacokinetics experiment, for an IgM antibody that has an HSA-15-J configuration on the J-chain. Panel B is a graph showing concentration as a function of time for a mouse PK experiment with IgM antibody that has a J-15-HSA configuration on the J-chain.

FIG. 22 is a table showing alpha and beta half-life in hours and AUC for 6 different antibodies.

FIG. 23 is an image of a reducing PAGE gel and a Western blot analysis of the antibodies listed in the table, one of which (1.5.3V15J15ABD) has bidentate J-chain configuration.

FIG. 24, is a graph showing CDC activity as a function of concentration for antibodies having the indicated J-chain configuration. The bidentate ABD-IgM has essentially the same activity as IgM with or without J-chain.

FIG. 25, is a graph showing CDC activity as a function of concentration for antibodies having the indicated J-chain configuration. The bidentate HSA-IgM has essentially the same activity as IgM with or without J-chain.

FIG. 26, Panel A, is a graph showing concentration as a function of time for an IgM antibody that has a V-J-ABD bidentate J-chain configuration. Panel B is a graph showing concentration as a function of time for an IgM antibody that has a V-J-HSA bidentate J-chain configuration.

FIG. 27 is a table showing alpha and beta half-life in hours and AUC parameters for 4 different antibodies with various configurations of their J-chains.

FIG. 28, Panel A and Panel B are graphs showing percentage of pre-dose CD19+ B-cells as a function of dose (ng/mouse) for various constructs (e.g., 1.5.3V15J15HSAwt and 1.5.3V15J15HSA (K573P)).

DETAILED DESCRIPTION OF THE INVENTION

I. Definitions

[0023] Where a range of values is provided, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limit of that range and any other stated or intervening value in that stated range is encompassed within the invention. The upper and lower limits of these smaller ranges may independently be included in the smaller ranges encompassed within the invention, subject to any specifically excluded limit in the stated range.

[0024] Unless defined otherwise, technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Singleton et al., Dictionary of Microbiology and Molecular Biology 2nd ed., J. Wiley & Sons (New York, NY 1994), provides one skilled in the art with a general guide to many of the terms used in the present application.

[0025] The term "ADME" as used herein is an abbreviation for absorption, distribution, metabolism, and excretion, and is used in the broadest sense to describe the disposition of a pharmaceutical compound within an organism.

[0026] The term "ADME-modulating moiety" is used herein in the broadest sense to encompass any chemical entity capable of modulating one or more of the absorption, distribution, metabolism and excretion characteristics of a molecule to which it is attached. Examples of ADME-modulating moieties include, without limitation, antibodies, antigen-binding fragments of antibodies, antibody-drug conjugates, antibody-like molecules, antigen-binding fragments of antibody-like molecules, ligands, receptors, proteins, and polypeptides (including peptides). Preferred binding moieties are antigen-binding fragments of antibodies, preferably with a biological function. An example of a biological function is the ability of an ADME-modulating moiety to bind to a target that extends the half-life of a subject binding molecule.

[0027] The term "antibody" includes monoclonal antibodies (including full length antibodies which have an immunoglobulin Fc region), single-chain molecules, as well as antibody fragments (e.g., Fab, $F(ab')_2$, and Fv). The term "immunoglobulin" (Ig) is used interchangeably with "antibody" herein. The basic 4-chain antibody unit is a heterotetrameric glycoprotein composed of two identical light (L) chains and two identical heavy (H) chains. Unless noted otherwise, the

term "antibody" is used herein in the broadest sense and specifically includes all isotypes, sub-classes and forms of antibodies, including IgG, IgM, IgA, IgD, and IgE antibodies and their fragments, preferably antigen-binding fragments. Preferred antibodies herein include IgM and IgA antibodies and their antigen-binding fragments, which may be modified to include sequences from other isotypes, such as IgG to produce chimeric antibodies.

[0028] In the case of IgGs, the 4-chain unit is generally about 150,000 daltons. Each L chain is linked to an H chain by one covalent disulfide bond, while the two H chains are linked to each other by one or more disulfide bonds depending on the H chain isotype. Each H and L chain also has regularly spaced intrachain disulfide bridges. Each H chain has at the N-terminus, a variable domain (V_H) followed by three constant domains (C_H) for each of the α and γ chains and four C_H domains for μ and ε isotypes. Each L chain has at the N-terminus, a variable domain (V_L) followed by a constant domain at its other end. The V_L is aligned with the V_H and the C_L is aligned with the first constant domain of the heavy chain (C_{H1}). Particular amino acid residues are believed to form an interface between the light chain and heavy chain variable domains. The pairing of a V_H and V_L together forms a single antigen-binding site.

[0029] IgM is a glycoprotein which forms polymers where multiple immunoglobulins are covalently linked together with disufide bonds. IgM mostly exists as a pentamer but also as a hexamer and therefore contains 10 or 12 antigen binding sites. The pentameric form typically contains an additional polypeptide, called the J-chain, but can also be made in the absence of J-chain. The pentameric IgM molecule has a molecular weight of approximately 970 kDa. Due to its polymeric nature, IgM possesses high avidity and is particularly effective in complement activation. Unlike in IgG, the heavy chain in IgM monomers is composed of one variable and four constant domains. The IgM constant domains are designated herein as CM1 or Cμ1, CM2 or Cμ2, CM3 or Cμ3, and CM4 or Cμ4, wherein the "CM" and Cμ" designations are used interchangeably. The structure of an IgM pentamer is illustrated in FIG. 1.

[0030] The term "IgM" is used herein in the broadest sense and specifically includes mono-, and multi-specific (including bispecific) IgM molecules, such as, for example, the multi-specific IgM binding molecules disclosed in PCT Publication No. WO2015053887A1.

[0031] The term "IgM binding unit" or "IgM antibody binding unit" is used in the broadest sense and specifically covers an IgM antibody heavy chain constant region polypeptide, comprising at least a CM4 constant domain, fused to a variable domain sequence (V_H) binding to a target (e.g., antigen), with or without an associated antibody light chain variable domain (V_L) sequence.

[0032] The term "bispecific IgM binding unit" or "bispecific IgM antibody binding unit" is used in the broadest sense and specifically covers a pair of IgM antibody heavy chain constant region polypeptides, comprising at least a CM4 constant domain, fused to a variable domain sequence (V_H), each variable domain sequence binding to a different target, with or without associated antibody light chain variable domain (V_L) sequences. In one embodiment, the bispecific IgM antibody comprises two V_HV_L antigen binding regions, each capable of binding to a different epitope on one antigen or epitopes on two different antigens. The bispecific IgM antibody binding units can be full length from a single species, or be chimerized or humanized. The bispecific IgM antibodies of the present invention have a penta- or hexameric ring structure comprising five or six bispecific IgM binding units.

[0033] The term "multi-specific IgM" is used herein in the broadest sense to refer to IgM antibodies with two or more binding specificities. Thus, the term "multi-specific" includes "bispecific", e.g., bispecific antibodies or bispecific binding units, including IgM pentamers comprising at least two monospecific subunits, each binding to a different antigen (AA, BB), or five or six bispecific subunits, each binding to two different antigens (AB, AB). Thus, the bispecific and multi-specific IgM pentamers may include five identical bispecific binding units, monospecific IgM binding units, at least two of them have different binding specificities, or any combination thereof.

[0034] A "full length IgM antibody heavy chain" is a polypeptide consisting in N-terminal to C-terminal direction of an antibody heavy chain variable domain (VH), an antibody constant heavy chain constant domain 1 (CM1 or Cμ1), an antibody heavy chain constant domain 2 (CM2 or Cμ2), an antibody heavy chain constant domain 3 (CM3 or Cμ3), and an antibody heavy chain constant domain 4 (CM4 or Cμ4). The bispecific full length IgM antibodies as defined herein comprise five or six monomers (binding units), each with two antigen binding sites, which specifically bind to two different binding targets (epitopes). The C-terminus of the heavy or light chain of the full length antibody denotes the last amino acid at the C-terminus of the heavy or light chain. The N-terminus of the heavy or light chain of the full length antibody denotes the first amino acid at the N-terminus of the heavy or light chain.

[0035] Native IgA is a tetrameric protein comprising two identical light chains (κ or λ) and two identical heavy chains (α). In the human, there are two IgA isotypes, IgA1 and IgA2. IgA, similarly to IgG, contains three constant domains (CA1-CA3 or Cα1-Cα3), with a hinge region between the Cα1 and Cα2 domains, wherein the "CA" and "Cα" designations are used interchangeably. All IgA isotypes have an 18 amino acid "tailpiece", which is located C-terminal to the Cα3 domain, which enables polymeric Ig formation (see, e.g., Garcia-Pardo et al., 1981, J. Biol. Chem. 256, 11734-11738 and Davis et al., 1988, Eur. J. Immunol. 18, 1001-1008). Serum IgA is a monomer but can also polymerize. In its secretory form IgA comprises from 2-5 of the basic 4-chain units, linked by a J-chain, which may include a tail-piece, and may be associated by a secretory component. The structures of tail-piece, dimeric IgA and secretory IgA, associated with a secretory component (sIgA) are illustrated in FIG. 2. IgA antibodies can be further divided into IgA1 and IgA2 sub-classes.

The term "IgA" antibody is used herein to specifically include all sub-classes, i.e., IgA1 and IgA2 antibodies, including dimeric and multimeric forms, with and without a secretory component, as well as fragments, preferably antigen-binding fragments, of such antibodies. For the purposes of the present invention, the IgA antibody preferably is a dimer, where two tail-pieces are connected by a J-chain (see, FIG. 2).

[0036] The term "IgA" is used herein in the broadest sense and specifically includes mono-, and multi-specific IgA molecules, such as, for example, the multi-specific IgA binding molecules disclosed in PCT Publication No. WO2015120474A1.

[0037] The term "multi-specific IgA" is used herein in the broadest sense to refer to IgA antibodies with two or more binding specificities. Thus, the term "multi-specific" includes "bispecific", e.g., bispecific antibodies or bispecific binding units, including IgA dimers comprising two monospecific subunits, each binding to a different antigen (AA, BB), or two bispecific subunits, each binding to two different antigens (AB, AB).

[0038] In one embodiment, the dimeric multi-specific IgA molecules consist of two monospecific binding units, each binding unit having binding specificity to a different binding target (AA, BB). In another embodiment, in the dimeric IgA molecules at least one of the two binding units has two different binding specificities (i.e., is a bispecific, e.g., AA, A,B or AA, BC). In another embodiment, each of the two binding units has two specificities, which may be the same (AB, AB) or different (AC, CD or AB, AC, for example).

[0039] The term "bispecific IgA antibody binding unit" is used in the broadest sense and specifically covers a pair of IgA antibody heavy chain constant region polypeptides, comprising at least a CA3 constant domain, fused to a variable domain sequence (V_H), each variable domain sequence binding to a different target, with or without associated antibody light chain variable domain (V_L) sequences. In one embodiment, the bispecific IgA antibody comprises two V_HV_L antigen binding regions, each capable of binding to a different epitope on one antigen or epitopes on two different antigens. The bispecific IgA antibody binding units can be full length from a single species, or be chimerized or humanized.

[0040] A "full length IgA antibody heavy chain" is a polypeptide consisting in N-terminal to C-terminal direction of an antibody heavy chain variable domain (VH), an antibody constant heavy chain constant domain 1 (CA1 or Cα1), an antibody constant heavy chain constant domain 2 (CA2 or Cα2), and an antibody heavy chain constant domain 3 (CA3 or Cα3). The bi- or multi-specific full length IgA antibodies according to the invention comprise two monomers (binding units), each of which may be mono- or bispecific, with or without a secretory component. Thus, the multi-specific IgA antibodies of the present invention may include monospecific and bispecific binding units, provided that the resultant IgA antibody has at least two binding specificities. The C-terminus of the heavy or light chain of the full length antibody denotes the last amino acid at the C-terminus of the heavy or light chain. The N-terminus of the heavy or light chain of the full length antibody denotes the first amino acid at the N-terminus of the heavy or light chain.

[0041] For further details of the structure and properties of the different classes of antibodies, see e.g., Basic and Clinical Immunology, 8th Edition, Daniel P. Stites, Abba I. Terr and Tristram G. Parslow (eds), Appleton & Lange, Norwalk, Conn., 1994, page 71 and Chapter 6.

[0042] The term "interface", as used herein, is used to refer to a region, which comprises those "contact" amino acid residues (or other non-amino acid groups such as, for example, carbohydrate groups,) in a first IgM heavy chain constant region which interact with one or more "contact" amino acid residues (or other non-amino acid groups) in a second IgM heavy chain constant region.

[0043] The term "asymmetric interface" is used to refer to an interface (as hereinabove defined) formed between two antibody chains, such as a first and a second IgM heavy chain constant region and/or between an IgM heavy chain constant region and its matching light chain, wherein the contact residues in the first and the second chains are different by design, comprising complementary contact residues. The asymmetric interface can be created by knobs/holes interactions and/or salt bridges coupling (charge swaps) and/or other techniques known in the art, such as for example, by the CrossMab approach for coupling a μ heavy chain to its matching light chain.

[0044] A "cavity" or "hole" refers to at least one amino acid side chain which is recessed from the interface of the second polypeptide and therefore accommodates a corresponding protuberance ("knob") on the adjacent interface of the first polypeptide. The cavity (hole) may exist in the original interface or may be introduced synthetically (e.g., by altering nucleic acid encoding the interface). Normally, nucleic acid encoding the interface of the second polypeptide is altered to encode the cavity. To achieve this, the nucleic acid encoding at least one "original" amino acid residue in the interface of the second polypeptide is replaced with DNA encoding at least one "import" amino acid residue which has a smaller side chain volume than the original amino acid residue. It will be appreciated that there can be more than one original and corresponding import residue. The upper limit for the number of original residues which are replaced is the total number of residues in the interface of the second polypeptide. The preferred import residues for the formation of a cavity are usually naturally occurring amino acid residues and are preferably selected from alanine (A), serine (S), threonine (T), valine (V) and glycine (G). Most preferred amino acid residues are serine, alanine or threonine, most preferably alanine. In the preferred embodiment, the original residue for the formation of the protuberance has a large side chain volume, such as tyrosine (Y), arginine (R), phenylalanine (F) or tryptophan (W).

[0045] An "original" amino acid residue is one which is replaced by an "import" residue which can have a smaller or

larger side chain volume than the original residue. The import amino acid residue can be a naturally occurring or non-naturally occurring amino acid residue, but preferably is the former.

[0046] By "non-naturally occurring" amino acid residue is meant a residue which is not encoded by the genetic code, but which is able to covalently bind adjacent amino acid residue(s) in the polypeptide chain. Examples of non-naturally occurring amino acid residues are norleucine, ornithine, norvaline, homoserine and other amino acid residue analogues such as those described in Ellman et al., Meth. Enzym. 202:301-336 (1991), for example. To generate such non-naturally occurring amino acid residues, the procedures of Noren et al. Science 244: 182 (1989) and Ellman et al., supra can be used. Briefly, this involves chemically activating a suppressor tRNA with a non-naturally occurring amino acid residue followed by in vitro transcription and translation of the RNA. The methods of the current invention, in certain embodiments, involve replacing at least one original amino acid residue in an IgM heavy chain, but more than one original residue can be replaced. Normally, no more than the total residues in the interface of the first or second polypeptide will comprise original amino acid residues which are replaced. The preferred original residues for replacement are "buried". By "buried" is meant that the residue is essentially inaccessible to solvent. The preferred import residue is not cysteine to prevent possible oxidation or mispairing of disulfide bonds.

[0047] The protuberance is "positionable" in the cavity which means that the spatial location of the protuberance and cavity on the interface of the first polypeptide and second polypeptide respectively and the sizes of the protuberance and cavity are such that the protuberance can be located in the cavity without significantly perturbing the normal association of the first and second polypeptides at the interface. Since protuberances such as Tyr, Phe and Trp do not typically extend perpendicularly from the axis of the interface and have preferred conformations, the alignment of a protuberance with a corresponding cavity relies on modeling the protuberance/cavity pair based upon a three-dimensional structure such as that obtained by X-ray crystallography or nuclear magnetic resonance (NMR). This can be achieved using widely accepted techniques in the art, including techniques of molecular modeling.

[0048] By "original nucleic acid" is meant the nucleic acid encoding a polypeptide of interest which can be "altered" (i.e., genetically engineered or mutated) to encode a protuberance or cavity. The original or starting nucleic acid may be a naturally occurring nucleic acid or may comprise a nucleic acid which has been subjected to prior alteration (e.g., a humanized antibody fragment). By "altering" the nucleic acid is meant that the original nucleic acid is mutated by inserting, deleting or replacing at least one codon encoding an amino acid residue of interest. Normally, a codon encoding an original residue is replaced by a codon encoding an import residue. Techniques for genetically modifying a DNA in this manner have been reviewed in Mutagenesis: a Practical Approach, M. J. McPherson, Ed., (IRL Press, Oxford, UK. (1991), and include site-directed mutagenesis, cassette mutagenesis and polymerase chain reaction (PCR) mutagenesis, for example.

[0049] The protuberance or cavity can be "introduced" into the interface of the first or second polypeptide by synthetic means, e.g., by recombinant techniques, in vitro peptide synthesis, those techniques for introducing non-naturally occurring amino acid residues previously described, by enzymatic or chemical coupling of peptides or some combination of these techniques. According, the protuberance or cavity which is "introduced" is "non-naturally occurring" or "non-native", which means that it does not exist in nature or in the original polypeptide (e.g., a humanized monoclonal antibody).

[0050] Preferably the import amino acid residue for forming the protuberance has a relatively small number of "rotamers" (e.g., about 3-6). A "rotamer" is an energetically favorable conformation of an amino acid side chain. The number of rotamers for the various amino acid residues are reviewed in Ponders and Richards, J. Mol. Biol. 193: 775-791 (1987).

[0051] Unless stated otherwise, the term "antibody" specifically includes native human and non-human IgG1, IgG2, IgG3, IgG4, IgE, IgA, IgD and IgM antibodies, including naturally occurring variants. Thus, for example, the human IgM sequence is available under GenBank Accession Number X14940.1, while variants have been reported as GenBank CAB37838.1, CAC20458.1, AFM37312.1, X57331.1, and J00260.1.

[0052] The term "native" with reference to a polypeptide (e.g., an antibody or a J-chain) is used herein to refer to a polypeptide having a sequence that occurs in nature, regardless of its mode of preparation. Thus, the terms "native" and "native sequence" are used herein interchangeably, and expressly encompass recombinant polypeptides with a sequence that is found in nature.

[0053] The term "native sequence J-chain" or "native J-chain" as used herein refers to J-chain of native sequence IgM or IgA antibodies of any animal species, including mature human J-chain, the amino acid sequence of which is shown in FIG. 3 (SEQ ID NO: 1).

[0054] The term "modified J-chain" is used herein to refer to variants of native sequence J-chain polypeptides comprising an extraneous ADME-modulating moiety introduced into the native sequence. The introduction can be achieved by any means, including direct or indirect fusion of an extraneous ADME-modulating moiety or by attachment through a chemical linker. The term "modified human J-chain" specifically encompasses, without limitation, a native sequence human J-chain of the amino acid sequence of SEQ ID NO: 1 modified by the introduction of an ADME-modulating moiety. The term specifically encompasses, without limitation, a native sequence human J-chain of the amino acid sequence of SEQ ID NO: 1 modified by the introduction of an extraneous ADME-modulating moiety which does not interfere with efficient polymerization (dimerization) of IgM or IgA and binding of such polymers (dimers) to a target

[0055] The term "polypeptide" is used herein in the broadest sense and includes peptide sequences. The term "peptide" generally describes linear molecular chains of amino acids containing up to about 60, preferably up to about 30 amino acids covalently linked by peptide bonds.

[0056] The term "monoclonal antibody" as used herein refers to an antibody obtained from a population of substantially homogeneous antibodies, i.e., the individual antibodies comprising the population are identical except for possible naturally occurring mutations that may be present in minor amounts. Monoclonal antibodies are highly specific, being directed against a single antigenic site. Furthermore, in contrast to conventional (polyclonal) antibody preparations which typically include different antibodies directed against different determinants (epitopes), each monoclonal antibody is directed against a single determinant on the antigen. The modifier "monoclonal" indicates the character of the antibody as being obtained from a substantially homogeneous population of antibodies, and is not to be construed as requiring production of the antibody by any particular method. For example, the monoclonal antibodies to be used in accordance with the present invention may be made by the hybridoma method first described by Kohler et al. (1975) Nature 256:495, or may be made by recombinant DNA methods (see, e.g., U.S. Patent No. 4,816,567). The "monoclonal antibodies" may also be isolated from phage antibody libraries using the techniques described in Clackson et al. (1991) Nature 352:624-628 and Marks et al. (1991) J. Mol. Biol. 222:581-597, for example.

[0057] The monoclonal antibodies herein specifically include "chimeric" antibodies (immunoglobulins) in which a portion of the heavy and/or light chain is identical with or homologous to corresponding sequences in antibodies derived from a particular species, while the remainder of the chain(s) is identical with or homologous to corresponding sequences in antibodies derived from another species, as well as fragments of such antibodies, so long as they exhibit the desired biological activity (U.S. Patent No. 4,816,567; and Morrison et al. (1984) Proc. Natl. Acad. Sci. USA 81:6851-6855).

[0058] "Humanized" forms of non-human (e.g., murine) antibodies are antibodies which contain minimal sequence derived from non-human immunoglobulin. For the most part, humanized antibodies are human immunoglobulins (recipient antibody) in which residues from a hypervariable region of the recipient are replaced by residues from a hypervariable region of a non-human species (donor antibody) such as mouse, rat, rabbit or nonhuman primate having the desired specificity, affinity, and capacity. In some instances, Fv framework region (FR) residues of the human immunoglobulin are also replaced by corresponding non-human residues. Furthermore, humanized antibodies may comprise residues which are not found in the recipient antibody or in the donor antibody. These modifications are made to further refine antibody performance. In general, the humanized antibody will comprise substantially all of at least one, and typically two, variable domains, in which all or substantially all of the hypervariable loops correspond to those of a non-human immunoglobulin and all or substantially all of the FR regions are those of a human immunoglobulin sequence. The humanized antibody optionally also will comprise at least a portion of an immunoglobulin constant region (Fc), typically that of a human immunoglobulin. For further details, see Jones et al. (1986) Nature 321:522-525; Riechmann et al. (1988) Nature 332:323-329; and Presta (1992) Curr. Op. Struct. Biol. 2:593-596.

[0059] An "isolated" antibody herein is one which has been identified and separated and/or recovered from a component of its natural environment in a recombinant host cell. Contaminant components of its natural environment are materials which would interfere with diagnostic or therapeutic uses for the antibody, and may include enzymes, hormones, and other proteinaceous or nonproteinaceous solutes, as well as undesired byproducts of the production. In a preferred embodiment, an isolated antibody herein will be purified (1) to greater than 95% by weight, or greater than 98% by weight, or greater than 99% by weight, as determined by SDS-PAGE or SEC-HPLC methods, (2) to a degree sufficient to obtain at least 15 residues of N-terminal or internal amino acid sequence by use of a amino acid sequencer, or (3) to homogeneity by SDS-PAGE under reducing or non-reducing conditions using Coomassie blue or, preferably, silver stain. Ordinarily, an isolated antibody will be prepared by at least one purification step.

[0060] The term "specific binding" or "specifically binds to" or is "specific for" refers to the binding of two members of a binding pair, such as the binding of an antibody to a target antigen, e.g., an epitope on a particular polypeptide, peptide, or other target (e.g., a glycoprotein target), and means binding that is measurably different from a non-specific interaction (e.g., a non-specific interaction may be binding to bovine serum albumin or casein). Specific binding can be measured, for example, by determining binding of a half-life extending moiety, or an antibody, or an antibody modified by introduction of a half-life extending moiety, to a target molecule compared to binding to a control molecule. For example, specific binding can be determined by competition with a control molecule that is similar to the target, for example, an excess of non-labeled target. In this case, specific binding is indicated if the binding of the labeled target to a probe is competitively inhibited by excess unlabeled target. The term "specific binding" or "specifically binds to" or is "specific for" a particular polypeptide or an epitope on a particular polypeptide target as used herein can be exhibited, for example, by a molecule having a Kd for the target of at least about 200 nM, alternatively at least about 150 nM, alternatively at least about 100 nM, alternatively at least about 60 nM, alternatively at least about 50 nM, alternatively at least about 40 nM, alternatively at least about 30 nM, alternatively at least about 20 nM, alternatively at least about 10 nM, alternatively at least about 8 nM, alternatively at least about 6 nM, alternatively at least about 4 nM, alternatively at least about 2 nM, alternatively at least about 1 nM, or greater. In certain instances, the term "specific binding" refers to binding where a molecule binds to a particular polypeptide or epitope on a particular polypeptide without substantially binding to any other polypeptide

or polypeptide epitope.

[0061] "Binding affinity" refers to the strength of the sum total of noncovalent 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 (Kd). For example, the Kd can be about 200 nM, 150 nM, 100 nM, 60 nM, 50 nM, 40 nM, 30 nM, 20 nM, 10 nM, 8 nM, 6 nM, 4 nM, 2 nM, 1 nM, or stronger. Affinity can be measured by common methods known in the art, including those described herein. Low-affinity antibodies generally bind antigen slowly and tend to dissociate readily, whereas high-affinity antibodies generally bind antigen faster and tend to remain bound longer. A variety of methods of measuring binding affinity are known in the art.

[0062] As used herein, the "Kd" or "Kd value" refers to a dissociation constant measured by a technique appropriate for the antibody and target pair, for example using surface plasmon resonance assays, for example, using a BIAcore™-2000 or a BIAcore™-3000 (BIAcore, Inc., Piscataway, N.J.) at 25°C. with immobilized antigen CM5 chips at about 10 response units (RU).

[0063] The terms "conjugate," "conjugated," and "conjugation" refer to any and all forms of covalent or non-covalent linkage, and include, without limitation, direct genetic or chemical fusion, coupling through a linker or a cross-linking agent, and non-covalent association.

[0064] The term "fusion" is used herein to refer to the combination of amino acid sequences of different origin in one polypeptide chain by in-frame combination of their coding nucleotide sequences. The term "fusion" explicitly encompasses internal fusions, i.e., insertion of sequences of different origin within a polypeptide chain, in addition to fusion to one of its termini. The term "fusion" is used herein to refer to the combination of amino acid sequences of different origin.

[0065] The term "valent" as used herein denotes the presence of a specified number of binding sites in an antibody. As such, the terms "bivalent", "tetravalent", and "hexavalent" denote the presence of two binding sites, four binding sites, and six binding sites, respectively. Thus, if in a bispecific IgA antibody according to the present invention each binding unit is bivalent, the bispecific IgA antibody will have 4 valencies.

[0066] The term "epitope" includes any molecular determinant capable of specific binding to an antibody. In certain embodiments, epitope determinants include chemically active surface groupings of molecules such as amino acids, sugar side chains, phosphoryl, or sulfonyl, and, in certain embodiments, may have specific three dimensional structural characteristics, and or specific charge characteristics. An epitope is a region of an antigen that is bound by an antibody. A "binding region" is a region on a binding target bound by a binding molecule.

[0067] "Polyepitopic specificity" refers to the ability to specifically bind to two or more different epitopes on the same or different target(s). "Monospecific" refers to the ability to bind only one epitope. According to one embodiment the bispecific IgM antibody binds to each epitope with an affinity of at least 10^{-7} M, or 10^{-8} M or better.

[0068] The term "target" or "binding target" is used in the broadest sense and specifically includes polypeptides, without limitation, nucleic acids, carbohydrates, lipids, cells, and other molecules with or without biological function as they exist in nature.

[0069] The term "antigen" refers to an entity or fragment thereof, which can bind to an antibody or trigger a cellular immune response. An immunogen refers to an antigen, which can elicit an immune response in an organism, particularly an animal, more particularly a mammal including a human. The term antigen includes regions known as antigenic determinants or epitopes, as defined above.

[0070] As used herein, the term "immunogenic" refers to substances that elicit the production of antibodies, and/or activate T-cells and/or other reactive immune cells directed against an antigen of the immunogen.

[0071] An "antigen-binding site" or "antigen-binding region" of an antibody of the present invention typically contains six complementarity determining regions (CDRs) which contribute in varying degrees to the affinity of the binding site for antigen. There are three heavy chain variable domain CDRs (CDRH1, CDRH2 and CDRH3) and three light chain variable domain CDRs (CDRL1, CDRL2 and CDRL3). The extent of CDR and framework regions (FRs) is determined by comparison to a compiled database of amino acid sequences in which those regions have been defined according to variability among the sequences and/or structural information from antibody/antigen complexes. Also included within the scope of the invention are functional antigen binding sites comprised of fewer CDRs (i.e., where binding specificity is determined by three, four or five CDRs). Less than a complete set of 6 CDRs may be sufficient for binding to some binding targets. Thus, in some instances, the CDRs of a VH or a VL domain alone will be sufficient. Furthermore, certain antibodies might have non-CDR-associated binding sites for an antigen. Such binding sites are specifically included within the present definition.

[0072] The term "host cell" as used in the current application denotes any kind of cellular system which can be engineered to generate the antibodies according to the current invention. In one embodiment Chinese hamster ovary (CHO) cells are used as host cells.

[0073] As used herein, the expressions "cell," "cell line," and "cell culture" are used interchangeably and all such designations include progeny. Thus, the words "transformants" and "transformed cells" include the primary subject cell

and cultures derived therefrom without regard for the number of transfers. It is also understood that all progeny may not be precisely identical in DNA content, due to deliberate or inadvertent mutations. Variant progeny that have the same function or biological activity as screened for in the originally transformed cell are included.

[0074] A nucleic acid is "operably linked" when it is placed in a functional relationship with another nucleic acid sequence. For example, DNA for a pre-sequence or secretory leader is operably linked to DNA for a polypeptide if it is expressed as a pre-protein that participates in the secretion of the polypeptide; a promoter or enhancer is operably linked to a coding sequence if it affects the transcription of the sequence; or a ribosome binding site is operably linked to a coding sequence if it is positioned so as to facilitate translation. Generally, "operably linked" means that the DNA sequences being linked are contiguous, and, in the case of a secretory leader, contiguous and in reading frame. However, enhancers do not have to be contiguous. Linking is accomplished by ligation at convenient restriction sites. If such sites do not exist, the synthetic oligonucleotide adaptors or linkers are used in accordance with conventional practice.

[0075] The term "extraneous" with reference to an "ADME-modulating moiety" is used herein to refer to an ADME-modulating moiety not present in a reference native polypeptide sequence at the same location. Thus, an extraneous polypeptide sequence (including peptide sequences), might be comprised within the corresponding native sequence but at a different location. In a preferred embodiment, the "extraneous" sequence is not present in the corresponding native sequence in any location. The term "antagonist" as used herein refers to a molecule that causes a decrease in a function or activity as compared to the same function or activity in the absence of the molecule. An "antagonist" of a signaling pathway is therefore a molecule whose presence causes a decrease in a function or activity of the signaling pathway. The term "antagonize" as used herein refers to causing a decrease in a function or activity.

[0076] The term "agonist" as used herein refers to a molecule that causes an increase in a function or activity as compared to the same function or activity in the absence of the molecule. An "agonist" of a signaling pathway is therefore a molecule whose presence causes an increase in a function or activity of the signaling pathway. The term "agonize" as used herein refers to causing an increase in a function or activity.

[0077] The term "T-cell inhibitory signaling pathway" as used herein refers to a T-cell signaling pathway that leads to a qualitative or quantitative decrease in, blocking or, or halting of a T-cell immune response.

[0078] The term "T-cell stimulatory signaling pathway" as used herein refers to a T-cell signaling pathway that leads to a qualitative or quantitative increase in or maintenance of a T-cell immune response.

[0079] The term "low level expression target" as used herein refers to a target whose expression level on a target cell ranges from 0 to 1+, as determined by immunohistochemistry (IHC) tissue analysis, preferably performed on frozen, formalin-fixed, paraffin-embedded tissue sections. Guidelines for determining expression level via IHC are provided, for example, by the College of American Pathologists (CAP), and are exemplified by the ASCO-CAP HER2 Test Guideline Recommendations, available at http://www.cap.org/apps/ docs/committees/immunohistochemistry/summary_of_recommendations.pdf.

[0080] The term "low affinity target" as used herein refers to a target whose binding interaction with an antibody has a dissociation constant K_d that is greater than or equal to a value ranging from about 10 to 100 nM, such as about 25 to about 75 nM, as measured by ELISA.

[0081] The term "half-life" is used herein in the broadest sense to refer to the period of time required for the concentration or amount of a binding molecule to be reduced by one-half in the body of a subject.

[0082] The term "albumin-binding polypeptide" as used herein refers to a polypeptide that specifically binds to an albumin protein.

[0083] The term "Fc domain" as used herein broadly refers to a C-terminal region of an immunoglobulin heavy chain, including native sequence Fc domains and variant Fc domains.

[0084] The terms "extra-vascular" and "extra-vascular space" as used herein broadly refer to a portion of a subject that is situated outside of the subject's blood vessels (e.g., arteries and veins).

[0085] The term "intra-articular space" as used herein refers to any portion of a subject that is situated inside of a joint that is located, e.g., between two bones (e.g., the inside of a knee joint).

[0086] The term "intra-vitreal space" as used herein refers to any portion of a subject that is situated inside of an eyeball.

DETAILED DESCRIPTION

Design and Production of Binding Molecules with Modified J-Chain

[0087] IgM is the first immunoglobulin produced by B cells in response to stimulation by antigen, and is present at around 1.5 mg/ml in serum with a half-life of 5 days. IgM is a pentameric or hexameric molecule. Just as IgG, IgM monomers consist of two light and two heavy chains. However, while IgG contains three heavy chain constant domains (C_H1, C_H2 and C_H3), the heavy (μ) chain of IgM additionally contains a fourth constant domain (C_H4), similarly to the ε heavy chains in IgE. This extra constant domain is located in place of the IgG and IgA proline-rich hinge region that is responsible for the rotational flexibility of the antigen-binding Fab domains relative to the Fc domain of IgG and IgA

antibodies.

[0088] Five IgM monomers form a complex with an additional small polypeptide chain (the J-chain) to form a native IgM molecule. The J-chain is considered to facilitate polymerization of μ chains before IgM is secreted from antibody-producing cells. While crystallization of IgM has proved to be notoriously challenging, Czajkowsky and Shao (PNAS 106(35): 14960-14965, 2009) recently published a homology-based structural model of IgM, based on the structure of the IgE Fc domain and the known disulfide pairings. The authors report that the human IgM pentamer is a mushroom-shaped molecule with a flexural bias. The IgM heavy (μ) chain contains five N-linked glycosylation sites: Asn-171, Asn-332, Asn-395, Asn-402 and Asn-563.

[0089] Immunoglobulin A (IgA), as the major class of antibody present in the mucosal secretions of most mammals, represents a key first line of defense against invasion by inhaled and ingested pathogens. IgA is also found at significant concentrations in the serum of many species, where it functions as a second line of defense mediating elimination of pathogens that have breached the mucosal surface. Receptors specific for the Fc region of IgA, FcαR, are key mediators of IgA effector function. Human IgA may have two different IgA heavy constant region (Cα) genes which give rise to the two subclasses, IgA1 and IgA2. The main difference between IgA1 and IgA2 resides in the hinge region that lies between the two Fab arms and the Fc region. IgA1 has an extended hinge region due to the insertion of a duplicated stretch of amino acids, which is absent in IgA2. IgA has the capacity to form dimers, in which two monomer units, each comprising two heavy chains and light chains, are postulated to be arranged in an end-to-end configuration stabilized by disulfide bridges and incorporation of a J-chain. Dimeric IgA, produced locally at mucosal sites, is transported across the epithelial cell boundary and out into the secretions by interaction with the polymeric immunoglobulin receptor (pIgR). During this process, the pIgR is cleaved and the major fragment, termed secretory component (SC), becomes covalently attached to the IgA dimer.

[0090] Both IgA and IgM possess an 18-amino acid extension in the C terminus called the "tail-piece" (tp). The IgM (μtp) and IgA (αtp) tail-pieces differ at seven amino acid positions. The IgM and IgA tail-piece is highly conserved among various animal species. The conserved penultimate cysteine residue in the IgA and IgM tail-pieces has been demonstrated to be involved in polymerization. Both tail-pieces contain an N-linked carbohydrate addition site, the presence of which is required for dimer formation in IgA and J-chain incorporation and pentamer formation in IgM. However, the structure and composition of the N-linked carbohydrates in the tail-pieces differ, suggesting differences in the accessibility of the glycans to processing by glycosyltransferases.

[0091] The nucleotide and/or protein sequences of J-chains of human, and various vertebrate animal species, such as cow, mouse, avian, amphibian, and rabbit, have been reported. The human J-chain contains eight cysteine residues, two (Cys13 and Cys69) are involved in disulfide bridges with the α or μ-chains (in IgA and IgM, respectively), and six are involved in intrachain disulfide bridges (Cys13: Cys101, Cys72: Cys92, Cys109: Cys134). The three-dimensional crystal structure of the J-chain has not been reported.

[0092] The binding molecules of the present invention include a J-chain that comprises an ADME-modulating moiety that modulates one or more ADME characteristics of the binding molecule, without interfering with the ability of the IgM, IgA, IgG/IgM or IgG/IgA antibody to bind to its binding target(s). A binding molecule can, for example, be an IgM antibody, an IgA antibody, or an IgG/IgM or IgG/IgA hybrid antibody, which contains an IgM or IgA tail-piece at the IgG heavy chain and thus combine the properties of IgG and IgA or IgA, including the ability to incorporate and form polymers with a modified J-chain whose ADME-modulating moiety modulates an ADME characteristic of the binding molecule. For further details on IgG/IgM and IgG/IgA hybrid antibodies see, e.g., Koteswara et al., Clinical Immunology 2001, 101(1):21-31. An illustration of an example binding molecule in accordance with aspects of the invention is depicted in FIG. 5. The depicted binding molecule comprises an IgM pentamer with binding specificity for a target antigen, and comprises an ADME-modulating moiety attached to the J-chain.

[0093] An ADME-modulating moiety in accordance with embodiments of the invention can include, without limitation, antibodies, antigen-binding fragments of antibodies, antibody-like molecules, antigen-binding fragments of antibody-like molecules, proteins, ligands and receptors. It is emphasized that any type of ADME-modulating moiety can be introduced into a J-chain, following the teaching of the present disclosure, by appropriately selecting the location and type of addition (e.g., direct or indirect fusion, chemical tethering, etc.).

[0094] In some embodiments, a binding molecule comprises an amino acid sequence listed in Table 10. In some embodiments, a binding molecule comprises an amino acid sequence that is substantially similar to an amino acid sequence listed in Table 10, for example, has at least about 80% amino acid sequence identity, alternatively, has about 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%. 99.5%, or about 99.9% amino acid sequence identity to an amino acid sequence that is listed in Table 10.

[0095] In a preferred embodiment, an ADME-modulating moiety comprises an antibody or an antigen-binding fragment of an antibody (also referred to as an "antibody fragment"), including monospecific, bispecific, and multi-specific antibodies and antibody fragments, that modulates an ADME characteristic of the binding molecule. The term "antibody fragment" is used in the broadest sense and includes, without limitation, Fab, Fab', $F(ab')_2$, scFab, scFv, and $(scFv)_2$ fragments, complementarity determining region (CDR) fragments, linear antibodies, single-chain antibody molecules, minibodies,

and multi-specific antibodies formed from antibody fragments. In a preferred embodiment, the antibody fragment is an scFv.

[0096] In another preferred embodiment, an ADME-modulating moiety comprises an antibody-like molecule, such as, for example, a human domain antibody (dAb), Dual-Affinity Re-Targeting (DART) molecule, a diabody, a di-diabody, dual-variable domain antibody, a Stacked Variable Domain antibody, a Small Modular ImmunoPharmaceutical (SMIP), a Surrobody, a strand-exchange engineered domain (SEED)-body, VHH (e.g., a camelid-like antibody molecule), or TandAb that functions by modulating an ADME characteristic of the binding molecule.

[0097] An ADME-modulating moiety can be introduced into a native J-chain sequence at any location that allows the ADME-modulating moiety to modulate an ADME characteristic of the binding molecule without interfering with the binding of the recipient IgM, IgA, IgG/IgM or IgG/IgA molecule to its binding target or binding targets. Preferred locations include at or near the C-terminus, at or near the N-terminus or at an internal location that, based on the three-dimensional structure of the J-chain, is accessible. In preferred embodiments, an ADME-modulating moiety is introduced into a native sequence J-chain within about 10 residues from the C-terminus or within about 10 amino acid residues from the N-terminus, where the native sequence J-chain preferably is human J-chain of SEQ ID NO: 1. In another embodiment, an ADME-modulating moiety is introduced into the native sequence human J-chain of SEQ ID NO: 1 in between cysteine residues 92 and 101 of SEQ ID NO: 1, or at an equivalent location of another native sequence J-chain. In a further embodiment, an ADME-modulating moiety is introduced into a native sequence J-chain, such as a J-chain of SEQ ID NO: 1, at or near a glycosylation site. Most preferably, an ADME-modulating moiety is introduced into the native sequence human J-chain of SEQ ID NO: 1 within about 10 amino acid residues from the C-terminus.

[0098] Introduction can be accomplished by direct or indirect fusion, i.e., by the combination of an ADME-modulating moiety amino acid sequences in one polypeptide chain by in-frame combination of their coding nucleotide sequences, with or without a peptide linker. The peptide linker (indirect fusion), if used, may, for example, be about 1 to 50, or about 1 to 40, or about 1 to 30, or about 1 to 20, or about 1 to 10, or about 10 to 20 amino acid residues, and may be present at one or both ends of an ADME-modulating moiety to be introduced into a J-chain sequence. In a preferred embodiment, the peptide linker is about 10 to 20, or 10 to 15 amino acids long. In another preferred embodiment, the peptide linker is 15 amino acids long.

[0099] An ADME-modulating moiety can also be appended to a native J-chain sequence by chemical linkage using heterobifunctional protein crosslinkers containing two different functional groups, which have their own reactivity and selectivity. These crosslinkers can be used in a one step process or can be used to create activated proteins, which can often be preserved and reacted with the second biomolecule in a separate step. Thus, for example, a heterobifunctional crosslinking reagent can be used to form conjugates between a J-chain and an ADME-modulating moiety. The reactive groups include, without limitation, imine reactive groups (such as NHS or sulfo-NHS), maleimide groups, and the like. Such crosslinkers, which can be cleavable or non-cleavable, have been used, for example, in the formation of hapten carrier proteins and in preparing enzyme-antibody conjugates. Chemically, the cleavable crosslinkers specifically include, without limitation, disulfide-based, hydrazone, and peptide linkers. A well-known and much studied enzyme-labile linker is a valine-citrulline linker, but other peptide linkers are also known and suitable. Typical representatives of non-cleavable linkers include thioethers, such as SMCC (*N*-succinimidyl-4-(*N*-maleimidomethyl)-cyclohexane-1-carboxylate). For further details see, e.g., Ducry L and Stump B, Bioconjugate Chem. 2010, 21:5-13. For listing of further suitable linkers see, e.g., Klein et al., Protein Engineering, Design & Selection; 2014, 27(10): 325-330.

[0100] In some embodiments, a modified J-chain comprises one extraneous ADME-modulating moiety. In some embodiments, a modified J-chain comprises more than one ADME-modulating moiety. For example, in some embodiments, one ADME-modulating moiety is introduced into a modified J-chain at either the N-terminus or the C-terminus. In some embodiments, a first ADME-modulating moiety is introduced into a modified J-chain at the N-terminus, and a second ADME-moiety is introduced into the same modified J-chain at the C-terminus. In some embodiments, an ADME-modulating moiety is introduced into a modified J-chain, and a binding moiety is introduced into the same modified J-chain. For examples, in some embodiments, an ADME-modulating moiety is introduced into a modified J-chain at the N-terminus, and a binding moiety (e.g., a CD3-binding antibody fragment, e.g., a CD3-binding scFv antibody fragment) is introduced into the same modified J-chain at the C-terminus. In some embodiments, an ADME-modulating moiety is introduced into a modified J-chain at the C-terminus, and a binding moiety (e.g., a CD3-binding antibody fragment, e.g., a CD3-binding scFv antibody fragment) is introduced into the same modified J-chain at the N-terminus. A binding molecule that comprises a binding moiety at both the N-terminus and the C-terminus of the J-chain is referred to herein as a binding molecule that comprises a "bidentate" J-chain.

[0101] The modified J-chain may be produced by well-known techniques of recombinant DNA technology, e.g., by expressing a nucleic acid encoding the modified J-chain in a suitable prokaryotic or eukaryotic host organism, such as CHO cells or E. coli. Thus, the modified J-chain may, for example, be expressed in E. coli, as described by Symersky et al., Mol Immunol 2000, 37:133-140.

[0102] In one embodiment, the J-chain can be initially modified by insertion of an enzyme recognition site, and post-translationally modified by a peptide or non-peptide linker, which can tether any extraneous ADME-modulating moiety

to the J-chain.

[0103] The modified J-chain can also be co-expressed with the heavy and light chains of the recipient IgM, IgA, IgG/IgM or IgG/IgA antibody. Although due to its complex structure, the large scale production of recombinant IgM has been difficult, several recombinant production systems for IgM using non-lymphoid cells have been reported, including co-expression of the IgM heavy (H) and light (L) chains in C6 glioma cells, CHO cells, and HeLa cells (see, e.g., WO89/01975 and Wood et al., J. Immunol. 145, 3011-3016 (1990) for expression in CHO cells). Expression of an IgM monoclonal antibody in E. coli, with or without a J-chain is described, e.g., in Azuma et al., Clin Cancer Res 2007, 13(9):2745-2750. Production of IgM in an immortalized human retina cell line expressing E1A and E1B proteins of an adenovirus is described in U. S. Application Publication No. 20060063234.

[0104] The recipient IgM, IgA, IgG/IgM or IgG/IgA antibody may be monospecific, bispecific or multi-specific. Bispecific and multi-specific IgM and IgA binding molecules, including antibodies, are described, for example, in PCT Publication No. WO2015053887A1 and WO2015120474A1.

[0105] A subject binding molecule can bind to any binding target via the IgM, IgA, IgG/IgM or IgG/IgA antibody, while an ADME-modulating moiety located on the J-chain modulates one or more ADME characteristics of the binding molecule. As such, the subject binding molecules can be used to provide high avidity binding to a target that is targeted by the IgM, IgA, IgG/IgM or IgG/IgA antibody, while the ADME-modulating moiety on the J-chain modulates one or more ADME characteristics of the binding molecule. Different types of ADME-modulating moieties are described herein, as are different classes of targets that can be targeted by an antibody portion of a subject binding molecule.

ADME-modulating moieties that reduce clearance

[0106] Aspects of the invention include binding molecules having an ADME-modulating moiety that reduces clearance of a binding molecule from the circulation of a subject, thereby increasing the half-life of the binding molecule in the subject. Albumin binding is known in the art as a general strategy for improving the pharmacokinetics of a protein. For example, non-covalent association with albumin has been shown to extend the half-life of short lived proteins. *E.g.,* Dennis, Mark S. et al., J. Biol. Chem., 2002, 277:35035-35043. As such, the use of albumin (human serum albumin), albumin-like proteins, albumin binding peptides, albumin binding antibody moieties (e.g., albumin binding scFv antibody fragments) as ADME-modulating moieties in a subject binding molecule provides an effective strategy for manipulating the pharmacokinetics of a binding molecule. In addition, the neonatal Fc receptor (FcRn) is known to provide a recycling pathway that provides immunoglobulin molecules with a longer circulating half-life. *E.g.,* Roopenian D.C. et al., Nature Reviews Immunology 7, 715-725 (2007). As such, the use of FcRn-binding proteins, Fc domains that bind to FcRn, or antibody moieties that bind to FcRn, also provide an effective strategy for manipulating the pharmacokinetics of a binding molecule. Without being held to theory, in some embodiments, ADME-modulating moieties that bind to FcRn provide an extended half-life by accessing an FcRn-mediated recycling pathway, rather than merely providing extended half-life due to an increase in the molecular weight of the binding compound.

[0107] In some embodiments, an ADME-modulating moiety comprises an albumin protein. Albumin proteins are soluble, non-glycosylated proteins that are commonly found in blood plasma. Albumin proteins are known to interact with the FcRn-mediated recycling pathway, and as a result, have an extraordinarily long circulatory half-life.

[0108] In certain embodiments, an ADME-modulating moiety binds to an albumin protein, thereby connecting itself to an albumin protein and taking advantage of the FcRn-mediated recycling pathway. As such, in certain embodiments, an ADME-modulating moiety comprises an albumin binding peptide. Non-limiting examples of albumin-binding peptides are described in US Patent Publication No. US20050287153. In some embodiments, an ADME-modulating moiety comprises an albumin-binding antibody moiety. Non-limiting examples of antibody moieties that bind to albumin include anti-albumin scFv, anti-albumin VHH, anti-albumin scFab, and anti-albumin dAb.

[0109] In some embodiments, an ADME-modulating moiety comprises an FcRn-binding peptide. In certain embodiments, an ADME-modulating moiety comprises an FcRn-binding antibody moiety. In some embodiments, an ADME-modulating moiety comprises an Fc domain of an immunoglobulin molecule that is bound by an FcRn receptor. Non-limiting examples of ADME-modulating moieties that reduce the clearance of a binding molecule are provided below in Table 1. Non-limiting examples of proteins that can be used to generate an antibody moiety that can be used as an ADME-modulating moiety in the subject binding molecules are provided in Table 1.

Table 1: Sequence information for ADME-modulating moieties

ADME-modulating moiety	Amino acid sequence information
albumin	GenBank Accession No.: NP_000468.1
albumin binding peptide	DLCLRDWGCLW (SEQ ID NO: 2)
albumin binding peptide	DICLPRWGCLW (SEQ ID NO: 3)

(continued)

ADME-modulating moiety	Amino acid sequence information
albumin binding peptide	MEDICLPRWGCLWGD (SEQ ID NO: 4)
albumin binding peptide	QRLMEDICLPRWGCLWEDDE (SEQ ID NO: 5)
albumin binding peptide	QGLIGDICLPRWGCLWGRSV (SEQ ID NO: 6)
albumin binding peptide	QGLIGDICLPRWGCLWGRSVK (SEQ ID NO: 7)
albumin binding peptide	EDICLPRWGCLWEDD (SEQ ID NO: 8)
albumin binding peptide	RLMEDICLPRWGCLWEDD (SEQ ID NO: 9)
albumin binding peptide	MEDICLPRWGCLWEDD (SEQ ID NO: 10)
albumin binding peptide	MEDICLPRWGCLWED (SEQ ID NO: 11)
albumin binding peptide	RLMEDICLARWGCLWEDD (SEQ ID NO: 12)
albumin binding peptide	EVRSFCTRWPAEKSCKPLRG (SEQ ID NO: 13)
albumin binding peptide	RAPESFVCYWETICFERSEQ (SEQ ID NO: 14)
albumin binding peptide	EMCYFPGICWM (SEQ ID NO: 15)
FcRn	GenBank Accession No.: P55899.1
Fc domain of IgG1	GenBank Accession No.: AAB24269.1
Fc domain of IgG2	GenBank Accession No.: AAR26706.1
Fc domain of IgG3	GenBank Accession No.: ACO54886.1
Fc domain of IgG4	GenBank Accession No.: AAG00912.1

ADME-modulating moieties that enhance penetration of the blood brain barrier

[0110] Aspects of the invention include binding molecules having an ADME-modulating moiety that enhances the ability of a binding molecule to penetrate the blood brain barrier of a subject, thereby increasing the concentration of the binding molecule in the brain extracellular fluid and central nervous system. The blood brain barrier is formed by brain endothelial cells, which are connected by tight junctions. The blood brain barrier permits selective transport of certain molecules into the brain extracellular fluid and the central nervous system, while denying passage to others.

[0111] Aspects of the invention include binding molecules having a moiety that binds to one or more targets in a receptor-mediated transcytosis (RMT) pathway, thereby facilitating transportation of a binding molecule across the blood brain barrier. Specific non-limiting examples of binding targets that are associated with an RMT pathway include: transferrin, transferrin receptor, insulin, insulin receptor, IGF-1, IGF-1 receptor, leptin, leptin receptor, basigin, Glut1 and CD98hc. RMT pathways are known in the art to facilitate passage of their respective ligands through the blood brain barrier and into the brain extracellular fluid and central nervous system of a mammalian subject. *E.g.,* Dennis et al., Neuropsychopharmacology Reviews (2012) 37, 302-303; Joy Yu Zuchero et al., Neuron 89, 70-82 (2016). As such, the use of RMT binding moieties (e.g., antibody moieties that bind to an RMT pathway target (e.g., an RMT-associated cell surface receptor and/or its associated ligand)) as ADME-modulating moieties in a subject binding molecule provides an effective strategy for enhancing penetration of the blood brain barrier and increasing the concentration of the binding molecule in the brain extracellular fluid and the central nervous system. Non-limiting examples of antibody moieties that can bind to an RMT pathway target include scFv, VHH, scFab, and dAb moieties.

[0112] In some embodiments, an ADME-modulating moiety comprises an antibody moiety that binds to a receptor in an RMT pathway. In some embodiments, an ADME-modulating moiety comprises an antibody moiety that binds to a ligand in an RMT pathway. In some embodiments, an ADME-modulating moiety comprises a ligand, or a portion of a ligand that is capable of binding to a receptor, in an RMT pathway (e.g., comprises a transferrin protein, or comprises at least a portion of a transferrin protein that is capable of binding to a transferrin receptor).

[0113] In some embodiments, an ADME-modulating moiety comprises a transferrin receptor-binding antibody moiety (e.g., a transferrin receptor-binding scFv). In some embodiments, an ADME-modulating moiety comprises a transferrin-binding antibody moiety (e.g., a transferrin-binding scFv). In certain embodiments, an ADME-modulating moiety comprises an insulin receptor-binding antibody moiety (e.g., an insulin receptor-binding scFv). In certain embodiments, an ADME-modulating moiety comprises an insulin-binding antibody moiety (e.g., an insulin-binding scFv). In certain em-

bodiments, an ADME-modulating moiety comprises an IGF-1 receptor-binding antibody moiety (e.g., an IGF-1 receptor-binding scFv). In certain embodiments, an ADME-modulating moiety comprises an IGF-1-binding antibody moiety (e.g., an IGF-1-binding scFv). In certain embodiments, an ADME-modulating moiety comprises a leptin receptor-binding antibody moiety (e.g., a leptin receptor-binding scFv). In certain embodiments, an ADME-modulating moiety comprises a leptin-binding antibody moiety (e.g., a leptin-binding scFv). In some embodiments, an ADME-modulating moiety comprises a basigin-binding antibody moiety (e.g., a basigin-binding scFv). In some embodiments, an ADME-modulating moiety comprises a Glut1-binding antibody moiety (e.g., a Glut1-binding scFv). In some embodiments, an ADME-modulating moiety comprises a CD98hc-binding antibody moiety (e.g., a CD98hc-binding scFv).

[0114] In some embodiments, an ADME-modulating moiety comprises a transferrin protein. In some embodiments, an ADME-modulating moiety comprises an IGF-1 protein. In some embodiments, an ADME-modulating moiety comprises a leptin protein. In some embodiments, an ADME-modulating moiety comprises a basigin protein. In some embodiments, an ADME-modulating moiety comprises a Glut1 protein. In some embodiments, an ADME-modulating moiety comprises a CD98hc protein. Non-limiting examples of proteins that can be used to generate an antibody moiety that can be used as an ADME-modulating moiety that enhances penetration of the blood brain barrier are provided in Table 2.

Table 2: Sequence information for ADME-modulating moieties

ADME-modulating moiety	GenBank Accession No.
Transferrin receptor	AAA61153.1
Insulin receptor	P06213.4
IGF-1 receptor	P08069.1
Leptin receptor	P48357.2
Transferrin	AAB22049.1
Leptin	AAH69452.1
Insulin	AAA59172.1
IGF-1	CAA01954.1
basigin	BAA08109.1
Glut1	P11166.2
CD98hc (4F2 cell-surface antigen heavy chain)	P08195.3

ADME-modulating moieties that increase half-life in extra-vascular spaces

[0115] Aspects of the invention include binding molecules having an ADME-modulating moiety that increases a half-life of a binding molecule in an extra-vascular space of a subject. Therapeutic proteins that are delivered directly to extra-vascular spaces such as intra-articular spaces, or intra-vitreal spaces, typically have a characteristically short half-life in the extra-vascular space. *E.g.,* Mordenti, J. et al., Toxicological Sciences 52, 101-106 (1999); Mordenti, J. et al., Toxicological Sciences 27(5), 536-544 (1999).

[0116] Hyaluronic acid is an anionic, non-sulfated glycosaminoglycan that is a major component of the extracellular matrix in certain extra-vascular spaces, such as intra-articular spaces and intra-vitreal spaces. As such, the use of compounds that bind to hyaluronic acid as ADME-modulating moieties provides an effective strategy for retaining a therapeutic molecule in such an extracellular space. The structure of hyaluronic acid is provided in FIG. 15.

[0117] Tumor necrosis factor-inducible gene 6 protein (TSG-6) is a 30 kDa secreted protein that contains a hyaluronan-binding domain. The hyaluronan-binding domain interacts with extracellular matrix in extra-vascular spaces and is involved with cell migration. As such, the use of TSG-6 as an ADME-modulating moiety provides an effective strategy for retaining a therapeutic molecule in an extracellular space.

[0118] In some embodiments, an ADME-modulating moiety comprises a hyaluronic acid binding protein (HABP). In some embodiments, an ADME-modulating moiety comprises a TSG-6 protein. In certain embodiments, an ADME-modulating moiety comprises a hyaluronic acid-binding antibody moiety. In certain embodiments, an ADME-modulating moiety comprises a TSG-6-binding antibody moiety. Non-limiting examples of antibody moieties include scFv, VHH, scFab, and dAb moieties. Non-limiting examples of ADME-modulating moieties that retain a binding molecule in an extracellular space are provided below in Table 3.

Table 3: Sequence information for ADME-modulating moieties

ADME-modulating moiety	Amino acid sequence information
Hyaluronic acid binding protein (HABP)	GenBank Accession No.: 2207280A
TSG-6	GenBank Accession No.: CAD13434.1

Antagonist Targets

[0119] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that antagonizes a T-cell inhibitory signaling pathway. T-cell inhibitory signaling pathways are known in the art, and include, without limitation, those described in *Pardoll* et al. Non-limiting examples of T-cell inhibitory signaling pathways and components thereof are described in further detail below.

[0120] One example of a T-cell inhibitory signaling pathway is the signaling pathway involving programmed cell death-1 (PD-1) and its ligand, programmed cell death ligand-1 (PD-L1). PD-1 is an inhibitory cell surface receptor protein of the immunoglobulin superfamily, and is involved in the regulation of T-cell function in immunity and self-tolerance. PD-L1 interacts with PD-1 on the surface of T-cells, and inhibits proliferation of T-cells by blocking cell cycle progression and cytokine production. *Id.*

[0121] Another example of a T-cell inhibitory signaling pathway is the signaling pathway involving T-cell immunoglobulin and mucin domain 3 (TIM3). TIM3 is a cell surface glycoprotein that is expressed on the surface of T-cells, and functions as an inhibitory molecule that is involved in the termination of Th1 cells. *Id.*

[0122] Another example of a T-cell inhibitory signaling pathway is the signaling pathway involving lymphocyte-activation gene 3 (LAG3). LAG3 belongs to the immunoglobulin superfamily, and functions as an inhibitor of cellular proliferation, activation and homeostasis of T-cells. *Id.*

[0123] As reviewed above, the subject binding molecules comprise a J-chain that comprises an ADME-modulating moiety. In some embodiments, an IgM, IgA, IgG/IgM or IgG/IgA antibody binds to a target that is involved in a T-cell inhibitory signaling pathway and antagonizes the inhibitory signaling pathway, thereby blocking or diminishing inhibitory signals that are received by a T-cell via the pathway, while the ADME-modulating moiety on the J-chain modulates an ADME characteristic of the binding molecule. Due to their higher avidity, the subject IgM, IgA, IgG/IgM or IgG/IgA antibodies act more effectively as antagonists when directed against T-cell inhibitory signaling pathway targets, as compared to IgG antibodies, which only have two binding sites. As a result, the T-cell's immune response is not blocked, halted or diminished, or, at least, the inhibition of the T-cell's immune response is reduced or diminished. The antibody of a subject binding molecule can be used to antagonize any T-cell inhibitory signaling pathway, including but not limited to the inhibitory signaling pathways that involve the proteins listed in Table 4, below. The GenBank Accession Numbers corresponding to the human protein sequences of these T-cell inhibitory signaling pathway targets are provided in Table 4, below.

Table 4: Sequence information for T-cell stimulatory signaling pathway targets

T-cell stimulatory signaling pathway member:	GenBank Accession No.
PD-1	AAC51773.1
PD-L1	Q9NZQ7.1
TIM3	AAL65158.1
LAG3	AAH52589.1

Agonist Targets

[0124] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that agonizes a T-cell stimulatory signaling pathway. T-cell stimulatory signaling pathways are known in the art, and include, without limitation, those described in *Pardoll* et al. Non-limiting examples of T-cell stimulatory signaling pathways and components thereof are described in further detail below.

[0125] CD137 is a member of the tumor necrosis factor receptor (TNF-R) superfamily, and is expressed on the surface of T-cells. Its function is to stimulate T-cell proliferation and cytokine secretion. *E.g., Pardoll* at 254. OX40 is another member of the tumor necrosis factor receptor superfamily that is expressed on T-cells, and it functions by delivering a stimulatory signal to T-cells that helps to maintain the immune response over time. *Id.*

[0126] Another T-cell stimulatory signaling pathway involves CD40. CD40 is a member of the tumor necrosis factor

receptor superfamily, and is expressed on antigen presenting cells. Engagement of CD40 with its ligand CD40L results in various T-cell stimulatory signals. *Id.*

[0127] Another T-cell stimulatory signaling pathway involves gluococorticoid-induced TNFR-related protein (GITR). GITR is a member of the tumor necrosis factor receptor superfamily, and is expressed on T-cells. It functions by increasing T-cell proliferation, activation and cytokine production. *E.g.,* Nocentini, G. et al., Proc Natl Acad Sci U S A. 1997 Jun 10; 94(12):6216-21.

[0128] CD27 is another protein that is involved in a T-cell stimulatory signaling pathway. Another member of the tumor necrosis factor receptor superfamily, CD27 is expressed on the surface of T-cells and functions by delivering a stimulatory signal to T-cells when it interacts with CD70. *E.g., Pardoll* at 254.

[0129] Another T-cell stimulatory signaling pathway involves herpesvirus entry mediator (HVEM). HVEM is a member of the tumor necrosis factor receptor superfamily, and is expressed on the surface of antigen presenting cells. When HVEM interacts with certain ligands, such as CD258, it delivers a stimulatory signal to T-cells. *Id.*

[0130] As reviewed above, the subject binding molecules comprise ADME-modulating moiety on the J-chain that modulates an ADME characteristic of the binding molecule. In some embodiments, an IgM, IgA, IgG/IgM or IgG/IgA antibody binds to a target that is involved in a T-cell stimulatory signaling pathway and agonizes the stimulatory signaling pathway, thereby maintaining or increasing stimulatory signals that are received by a T-cell via the pathway, while the ADME-modulating moiety on the J-chain modulates an ADME characteristic of the binding molecule. Due to their higher avidity, the subject IgM, IgA, IgG/IgM or IgG/IgA antibodies act more effectively as agonists when directed against T-cell stimulatory signaling pathway targets, as compared to IgG antibodies, which only have two binding sites. As a result, a T-cell's immune response is maintained or increased. An antibody of a subject binding molecule can be used to agonize any T-cell stimulatory signaling pathway, including but not limited to the stimulatory signaling pathways that involve the proteins listed in Table 5, below. The GenBank Accession Numbers corresponding to the human protein sequences of these T-cell stimulatory signaling pathway targets are provided in Table 5, below.

Table 5: Sequence information for T-cell stimulatory signaling pathway targets

T-cell stimulatory signaling pathway member:	GenBank Accession No.
CD137 (4-1BB)	NP_001552.2
OX40	CAE11757.1
CD40	P25942.1
GITR	Q9Y5U5.1
CD27	P26842.2
HVEM	AAQ89238.1

[0131] Other non-limiting examples of T-cell stimulatory signaling pathways include those mediated by: TNFR1 (DR1) (GenBank Accession No. P19438.1); TNFR2 (GenBank Accession No. P20333.3); Fas (CD95, Apo1, DR2) (GenBank Accession No. AAH12479.1); CD30 (GenBank Accession No. AAA51947.1); TRAILR1 (DR4, Apo2) (GenBank Accession No. O00220.3); DR5 (TRAILR2) (GenBank Accession No. 014763.2); TRAELR3 (DcR1) (GenBank Accession No. 014798.3); TRAILR4 (DcR2) (GenBank Accession No. Q9UBN6.1); OPG (OCIF) (GenBank Accession No. O00300.3); TWEAKR (FN14) (GenBank Accession No. Q9NP84.1); DcR3 (GenBank Accession No. 095407.1); DR3 (GenBank Accession No. AAQ88676.1); EDAR (GenBank Accession No. Q9UNE0.1); and XEDAR (GenBank Accession No. AAQ89952.1). *See, e.g.,* Aggarwal et al., Blood, 119:651-665, 2012. In some embodiments, an IgM, IgA, IgG/IgM or IgG/IgA antibody binds to any one of these targets and agonizes a T-cell stimulatory signaling pathway, thereby maintaining or increasing stimulatory signals that are received by a T-cell via the pathway, while the ADME-modulating moiety on the J-chain modulates an ADME characteristic of the binding molecule.

Low Level Expression Targets

[0132] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that binds to a low level expression target. Due to their higher avidity, the subject binding molecules are more potent than IgG antibodies. As such, the subject binding molecules can be employed in settings where a particular binding target is expressed at a low level, and where higher avidity is beneficial in facilitating binding between an antibody and a target. An antibody of a subject binding molecule can be used to target any low level expression target. Specific examples of low level expression targets that may be targeted by an IgM, IgA, IgG/IgM or IgG/IgA antibody of the subject binding molecules include, without limitation, EGFR, HER2, HER3, EpCAM, CEACAM, Gp100, MAGE1 and PD-L1. The GenBank

Accession Numbers corresponding to the human protein sequences of these targets are provided in Table 6, below.

Table 6: Sequence information for low level expression targets

Target Name	GenBank Accession No.
EGFR	AAI18666.1
HER2	P04626.1
HER3	P21860.1
EpCAM	P16422.2
CEACAM	P06731.3
Gp100	AAC60634.1
MAGE1	NP_004979.3
PD-L1	Q9NZQ7.1

Low Affinity Targets

[0133] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that binds to a low affinity target. Due to their higher avidity, the subject binding molecules are more potent than IgG antibodies. As such, the subject binding molecules can be employed in settings where a particular binding target has a low binding affinity, and where higher avidity is beneficial in facilitating binding between an antibody and a target. An antibody of a subject binding molecule can be used to target any low affinity target. Specific examples of low affinity targets that may be targeted by an IgM, IgA, IgG/IgM or IgG/IgA antibody of the subject binding molecules include, without limitation, NY-ESO-1, Sialyl Lewis X antigen, and Tn antigen. The GenBank Accession Numbers corresponding to the human protein sequences of NY-ESO-1 and Sialyl Lewis X antigen are provided in Table 7, below. The structure of Tn antigen is provided in FIG. 14.

Table 7: Sequence information for low affinity targets

Target Name	GenBank Accession No.
NY-ESO-1	CAA05908.1
Sialyl Lewis X antigen	NP_001241688.1

Hematologic Cancer Targets

[0134] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that binds to a hematologic cancer target. Due to their higher avidity, the subject binding molecules are more potent than IgG antibodies. As such, the subject binding molecules can be employed in settings where a particular binding target is expressed at a low level, as is the case in certain hematologic cancers. The higher avidity of the subject binding molecules facilitates binding between an antibody and a target. An antibody of a subject binding molecule can be used to target any binding target, such as a low level expression target on a hematologic cancer cell. Specific examples of hematologic cancer targets that can be targeted by an IgM, IgA, IgG/IgM or IgG/IgA antibody of the subject binding molecules include, without limitation, CD19, CD20, CD22, CD33, CD38, CD52 and CD70. The GenBank Accession Numbers corresponding to the human protein sequences of these targets are provided in Table 8, below.

Table 8: Sequence information for hematologic cancer targets

Target Name	GenBank Accession No.
CD19	AAA69966.1
CD20	NP_690605.1
CD22	P20273.2
CD33	P20138.2

(continued)

Target Name	GenBank Accession No.
CD38	BAA18966.1
CD52	AJC 19276.1
CD70	NP_001243.1

Other binding targets

[0135] Aspects of the invention include binding molecules having an IgM, IgA, IgG/IgM or IgG/IgA antibody that binds to a target that is associated with a particular disease or disorder. Due to their higher avidity, the subject binding molecules are more potent than IgG antibodies. As such, the subject binding molecules can be employed in settings where high avidity binding to a particular binding target is desirable. An antibody of a subject binding molecule can be used to target any binding target. Specific examples of binding targets that can be targeted by an IgM, IgA, IgG/IgM or IgG/IgA antibody of the subject binding molecules include, without limitation, VEGF, TNF-alpha, amyloid beta, and Beta-secretase 1 (BACE) proteins. The GenBank Accession Numbers corresponding to the human protein sequences of these targets are provided in Table 9, below.

Table 9: Sequence information for other binding targets

Target Name	GenBank Accession No.
VEGF	AAP86646.1
TNF alpha	CAA26669.1
Amyloid beta A4	P05067.3
BACE (Beta-secretase 1)	P56817.2

Applications of Binding Molecules with ADME-modulating moieties

[0136] Binding molecules comprising a modified J-chain that comprises an ADME-modulating moiety have widespread therapeutic and diagnostic applications, including but not limited to the treatment of various diseases by modulating one or more ADME characteristics of a binding molecule.

[0137] In some embodiments, the subject binding molecules comprising a modified J-chain may broadly be used for the treatment of any of a variety of cancers. It is anticipated that any type of tumor and any type of tumor-associated antigen may be targeted by the subject binding molecules. Examples of cancer types include, without limitation, acute lymphoblastic leukemia, acute myelogenous leukemia, biliary cancer, breast cancer, cervical cancer, chronic lymphocytic leukemia, chronic myelogenous leukemia, colorectal cancer, endometrial cancer, esophageal, gastric, head and neck cancer, Hodgkin's lymphoma, lung cancer, medullary thyroid cancer, non-Hodgkin's lymphoma, multiple myeloma, renal cancer, ovarian cancer, pancreatic cancer, glioma, melanoma, liver cancer, prostate cancer, and urinary bladder cancer. However, the skilled artisan will realize that tumor-associated antigens are known in the art for virtually any type of cancer.

[0138] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that reduces clearance of the binding molecule from the circulation of a subject, while the antibody antagonizes a T-cell inhibitory signaling pathway. Without being held to theory, the purpose of such a binding molecule is to increase the half-life of the binding molecule via the J-chain ADME-modulating moiety, while simultaneously blocking or decreasing T-cell inhibitory signaling via the antibody. Due to their increased avidity, the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies act as effective antagonists when directed to certain binding targets, such as members of a T-cell inhibitory signaling pathway, as described above. Such binding molecules find utility, for example, in the treatment of diseases wherein blocking or decreasing the inhibition of a T-cell immune response is desirable, such as, e.g., certain cancers and immune disorders. Such cancers include, but are not limited to, epithelial cancers as well as hematologic cancers.

[0139] Epithelial cancers that are suitable for treatment with the subject binding molecules having an antagonist antibody and an ADME-modulating moiety on the J-chain include, without limitation, melanoma, non-small-cell lung, nasopharyngeal, colorectal, liver, urinary bladder, ovarian, gastric, esophageal, pancreatic, renal, thyroid or breast cancer, hormone receptor negative breast cancer, or triple negative breast cancer. Hematologic cancers that are suitable for treatment with the subject binding molecules having an antagonist antibody and an ADME-modulating moiety on the

J-chain include, without limitation, leukemia, lymphoma, myeloma, myelodysplastic syndrome, acute myeloid leukemia, acute lymphoblastic leukemia, chronic myelogenous leukemia, chronic lymphocytic leukemia, Hodgkin's lymphoma and non-Hodgkin's lymphoma. In some embodiments, the subject binding molecules are for use in the treatment of any of these conditions.

[0140] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that enhances penetration of the blood brain barrier by the binding molecule, while the antibody antagonizes a T-cell inhibitory signaling pathway. Without being held to theory, the purpose of such a binding molecule is to increase the concentration of the binding molecule in the brain extracellular fluid and the central nervous system via the J-chain ADME-modulating moiety, while simultaneously blocking or decreasing T-cell inhibitory signaling via the antibody. Due to their increased avidity, the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies act as effective antagonists when directed to certain binding targets, such as members of a T-cell inhibitory signaling pathway, as described above. Such binding molecules find utility, for example, in the treatment of diseases wherein blocking or decreasing the inhibition of a T-cell immune response is desirable, such as, e.g., certain cancers and immune disorders of the brain and central nervous system. Such cancers include, but are not limited to, glioma, astrocytoma, meningioma, neuroma and oligodendroglioma.

[0141] In some embodiments, the J-chain of the subject binding molecules includes an ADME-modulating moiety that reduces clearance of the binding molecule from the circulation of a subject, while the antibody agonizes a T-cell stimulatory signaling pathway. Without being held to theory, the purpose of such a binding molecule is to increase the half-life of the binding molecule via the ADME-modulating moiety on the J-chain, while simultaneously maintaining or increasing T-cell stimulatory signaling via the antibody. Due to their increased avidity, the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies act as super agonists when directed to certain binding targets, such as members of a T-cell stimulatory signaling pathway, as described above. Such binding molecules find utility, for example, in the treatment of diseases wherein maintenance or activation of a T-cell immune response is desirable, such as, e.g., certain cancers and immune disorders. Such cancers include, but are not limited to, epithelial cancers as well as hematologic cancers.

[0142] Epithelial cancers that are suitable for treatment with the subject binding molecules having an agonist antibody and an ADME-modulating moiety include on the J-chain include, without limitation, melanoma, non-small-cell lung, nasopharyngeal, colorectal, liver, urinary bladder, ovarian, gastric, esophageal, pancreatic, renal, thyroid or breast cancer, hormone receptor negative breast cancer, or triple negative breast cancer. Hematologic cancers that are suitable for treatment with the subject binding molecules having an agonist antibody and an ADME-modulating moiety include on the J-chain include, without limitation, leukemia, lymphoma, myeloma, myelodysplastic syndrome, acute myeloid leukemia, acute lymphoblastic leukemia, chronic myelogenous leukemia, chronic lymphocytic leukemia, Hodgkin's lymphoma and non-Hodgkin's lymphoma. In some embodiments, the subj ect binding molecules find use in the treatment of any of these conditions.

[0143] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that enhances penetration of the blood brain barrier by the binding molecule, while the antibody agonizes a T-cell stimulatory signaling pathway. Without being held to theory, the purpose of such a binding molecule is to increase the concentration of the binding molecule in the brain extracellular fluid and the central nervous system via the ADME-modulating moiety on the J-chain, while simultaneously maintaining or increasing T-cell stimulatory signaling via the antibody. Due to their increased avidity, the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies act as super agonists when directed to certain binding targets, such as members of a T-cell stimulatory signaling pathway, as described above. Such binding molecules find utility, for example, in the treatment of diseases wherein maintenance or activation of a T-cell immune response is desirable, such as, e.g., certain cancers and immune disorders of the brain and central nervous system. Such cancers include, but are not limited to, glioma, astrocytoma, meningioma and oligodendroglioma.

[0144] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that increases the half-life of the binding molecule, while the antibody binds to a low level expression target. Without being held to theory, the purpose of such a binding molecule is to increase the half-life of the binding molecule via the ADME-modulating moiety on the J-chain, while simultaneously binding to a low level expression target using the higher avidity of the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies. Such binding molecules find utility in the treatment of diseases wherein high avidity binding to a low level expression target is beneficial, such as, for example, in certain cancers and immune disorders. For example, certain epithelial cancers are known to express tumor antigens that have a low level of expression, as described above. Such epithelial cancers include, without limitation, melanoma, non-small-cell lung, nasopharyngeal, colorectal, liver, urinary bladder, ovarian, gastric, esophageal, pancreatic, renal, thyroid or breast cancer, hormone receptor negative breast cancer, or triple negative breast cancer. In some embodiments, the subject binding molecules are for use in the treatment of any of these conditions.

[0145] In some embodiments, the J-chain of the subject binding molecules includes an ADME-modulating moiety that increases the half-life of the binding molecule, while the antibody binds to a low affinity target. Without being held to theory, the purpose of such a binding molecule is to increase the half-life of the binding molecule via the ADME-modulating moiety on the J-chain, while simultaneously binding to a low affinity target using the higher avidity of the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies. As reviewed above, due to their increased avidity, the subject IgM, IgA, IgG/IgM

and IgG/IgA antibodies, comprising a modified J-chain comprising an ADME-modulating moiety are especially advantageous in situations where IgG antibodies bind to their target with low affinity. Thus, in some embodiments, the IgM, IgA, IgG/IgM and IgG/IgA antibodies herein may comprise the binding domain of a therapeutic IgG antibody. Such binding molecules find utility in the treatment of diseases wherein high avidity binding to a low affinity target is beneficial, such as, for example, in certain cancers and immune disorders. For example, certain epithelial cancers are known to express tumor antigens that have a low binding affinity, as described above. Such epithelial cancers include, without limitation, melanoma, non-small-cell lung, nasopharyngeal, colorectal, liver, urinary bladder, ovarian, gastric, esophageal, pancreatic, renal, thyroid or breast cancer, hormone receptor negative breast cancer, or triple negative breast cancer. In some embodiments, the subject binding molecules are for use in the treatment of any of these conditions.

[0146] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that increases the half-life of the binding molecule, while the antibody binds to a target on a hematologic cancer cell. Without being held to theory, the purpose of such a binding molecule is to increase the half-life of the binding molecule via the ADME-modulating moiety on the J-chain, while simultaneously binding to a hematologic cancer target using the higher avidity of the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies. Such binding molecules find utility in the treatment of hematologic cancers wherein high avidity binding to a tumor antigen is beneficial. For example, certain hematologic cancers are known to express tumor antigens at a low level, as described above. Such hematologic cancers include, without limitation, leukemia, lymphoma, myeloma, myelodysplastic syndrome, acute myeloid leukemia, acute lymphoblastic leukemia, chronic myelogenous leukemia, chronic lymphocytic leukemia, Hodgkin's lymphoma and non-Hodgkin's lymphoma. In some embodiments, the subject binding molecules are for use in the treatment of any of these conditions.

[0147] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that increases the retention of the binding molecule in an extra-vascular space, while the antibody binds to a binding target in the extra-vascular space. Without being held to theory, the purpose of such a binding molecule is to increase the residence time of the binding molecule in the extra-vascular space via the ADME-modulating moiety on the J-chain, while simultaneously binding to a binding target using the higher avidity of the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies. Such binding molecules find utility in the treatment of diseases or disorders wherein high avidity binding to a binding target in an extra-vascular space is beneficial. For example, tumor necrosis factor alpha (TNF alpha) is a binding target in the treatment of rheumatoid arthritis, which is an autoimmune disease that affects the joints of a subject. The subject binding molecules find use in the treatment of rheumatoid arthritis by providing high avidity binding to TNF alpha via the IgM, IgA, IgG/IgM or IgG/IgA antibody, while also providing extended retention time within an intra-articular space via the ADME-modulating moiety on the modified J-chain.

[0148] In another non-limiting example, vascular endothelial growth factor (VEGF) is a binding target in the treatment of age-related macular degeneration (AMD), which is a disease that affects the retina of a subject. The subject binding molecules find use in the treatment of AMD by providing high avidity binding to VEGF via the IgM, IgA, IgG/IgM or IgG/IgA antibody, while also providing extended retention time in an intra-vitreal space via the ADME-modulating moiety on the modified J-chain.

[0149] In some embodiments, the J-chain of a subject binding molecule includes an ADME-modulating moiety that enhances penetration of the blood brain barrier by the binding molecule, while the antibody binds to a binding target in the brain extracellular fluid or a central nervous system tissue. Without being held to theory, the purpose of such a binding molecule is to increase the concentration of the binding molecule in the brain extracellular fluid and central nervous system tissue via the ADME-modulating moiety on the J-chain, while simultaneously binding to a binding target using the higher avidity of the subject IgM, IgA, IgG/IgM and IgG/IgA antibodies. Such binding molecules find utility in the treatment of diseases or disorders wherein high avidity binding to a binding target in the brain extracellular fluid or central nervous system tissue is beneficial. For example, amyloid beta is a binding target in the treatment of Alzheimer's disease, which is a disease that affects the central nervous system of a subject. Beta secretase 1 (BACE) is also a binding target in the treatment of Alzheimer's disease. The subject binding molecules find use in the treatment of Alzheimer's disease by providing high avidity binding to, e.g., amyloid beta or BACE via the IgM, IgA, IgG/IgM or IgG/IgA antibody, while also providing increased concentration of the binding molecule within the brain extracellular fluid or central nervous system tissue via the ADME-modulating moiety on the modified J-chain.

[0150] Examples of IgM, IgA, IgG/IgM, or IgG/IgA antibodies including a modified J-chain that modulates an ADME characteristic of a binding molecule can include the binding regions of known IgG antibodies to tumor-associated antigens, such as, for example, blinatumomab (also known as MT103) (anti-CD19), CD19hA19 (anti-CD19, U.S. Pat. No. 7,109,304), hPAM4 (anti-mucin, U.S. Pat. No. 7,282,567), hA20 (anti-CD20, U.S. Pat. No. 7,251,164), hIMMU31 (anti-AFP, U.S. Pat. No. 7,300,655), hLL1 (anti-CD74, U.S. Pat. No. 7,312,318), hLL2 (anti-CD22, U.S. Pat. No. 7,074,403), hMu-9 (anti-CSAp, U.S. Pat. No. 7,387,773), hL243 (anti-HLA-DR, U.S. Pat. No. 7,612,180), hMN-14 (anti-CEACAM5, U.S. Pat. No. 6,676,924), hMN-15 (anti-CEACAM6, U.S. Pat. No. 7,541,440), hRS7 (anti-EGP-1, U.S. Pat. No. 7,238,785), hMN-3 (anti-CEACAM6, U.S. Pat. No. 7,541,440), Ab124 and Ab125 (anti-CXCR4, U.S. Pat. No. 7,138,496).

[0151] Other antibodies that can provide binding regions for use in combination with a modified J-chain that increases the half-life of a subject binding molecule include, for example, abciximab (anti-glycoprotein IIb/IIIa), alemtuzumab (anti-

CD52), bevacizumab (anti-VEGF), cetuximab (anti-EGFR), gemtuzumab (anti-CD33), ibritumomab (anti-CD20), panitumumab (anti-EGFR), tositumomab (anti-CD20), trastuzumab (anti-ErbB2), lambrolizumab (anti-PD-1 receptor), nivolumab (anti-PD-1 receptor), ipilimumab (anti-CTLA-4), abagovomab (anti-CA-125), adecatumumab (anti-EpCAM), atlizumab (anti-IL-6 receptor), benralizumab (anti-CD125), obinutuzumab (GA101, anti-CD20), CC49 (anti-TAG-72), AB-PG1-XG1-026 (anti-PSMA, U.S. patent application Ser. No. 11/983,372, deposited as ATCC PTA-4405 and PTA-4406), D2/B (anti-PSMA, WO 2009/130575), tocilizumab (anti-IL-6 receptor), basiliximab (anti-CD25), daclizumab (anti-CD25), efalizumab (anti-CD11a), GA101 (anti-CD20; Glycart Roche), atalizumab (anti-.alpha.4 integrin), omalizumab (anti-IgE); anti-TNF-.alpha. antibodies such as CDP571 (Ofei et al., 2011, Diabetes 45:881-85), MTNFAI, M2TNFAI, M3TNFAI, M3TNFABI, M302B, M303 (Thermo Scientific, Rockford, Ill.), infliximab (Centocor, Malvern, Pa.), certolizumab pegol (UCB, Brussels, Belgium), anti-CD40L (UCB, Brussels, Belgium), adalimumab (Abbott, Abbott Park, Ill.), BENLYSTA.RTM. (Human Genome Sciences); antibodies for therapy of Alzheimer's disease such as Alz 50 (Ksiezak-Reding et al., 1987, J Biol Chem 263:7943-47), gantenerumab, solanezumab and infliximab; anti-fibrin antibodies like 59D8, T2G1s, MH1; anti-CD38 antibodies such as MOR03087 (MorphoSys AG), MOR202 (Celgene), HuMax-CD38 (Genmab) or daratumumab (Johnson & Johnson); trastuzumab (anti-HER2); tremelimumab (anti-CTLA4); urelumab (anti-CD137 (4-1BB)); vorsetuzumab (anti-CD70); duligotumab (anti-HER3); dacetuzumab (anti-CD40); varlilumab (anti-CD27); atezolizumab (anti-PD-L1); anti-MAGE1 antibodies such as MA454 (Thermo Scientific, Rockford, IL); anti-OX-40 antibodies such as ACT35 (Affymetrix eBioscience, San Diego, CA); anti-GITR antibodies such as 621 (BioLegend, San Diego, CA); anti-HVEM antibodies such as 122 (BioLegend, San Diego, CA); anti-TIM3 antibodies such as F38-2E2 (BioLegend, San Diego, CA); anti-LAG3 antibodies such as 3DS223H (Affymetrix eBioscience, San Diego, CA); anti-BTLA antibodies such as MIH26 (BioLegend, San Diego, CA); anti-VISTA antibodies such as MAB71261 (R&D Systems, Minneapolis, MN); anti-TIGIT antibodies such as MBSA43 (Affymetrix eBioscience, San Diego, CA); anti-CEACAM antibodies such as D14HD11 (abcam, Cambridge, MA); anti-Gp100 antibodies such as ab52058 (abcam, Cambridge, MA); anti-NY-ESO-1 antibodies such as E978 (Thermo Scientific, Rockford, IL); anti-Sialyl Lewis X antigen antibodies such as MAB2096 (EMD Millipore, Billerica, MA); anti-Tn antigen antibodies such as MA1-90544 (Thermo Scientific, Rockford, IL); anti-HIV antibodies such as P4/D10 (U.S. Pat. No. 8,333,971), Ab 75, Ab 76, Ab 77 (Paulik et al., 1999, Biochem Pharmacol 58:1781-90), as well as the anti-HIV antibodies described in U.S. Pat. No. 5,831,034, U.S. Pat. No. 5,911,989, and Vcelar et al., AIDS 2007; 21(16):2161-2170 and Joos et al., Antimicrob. Agents Chemother. 2006; 50(5):1773-9; anti-albumin antibodies such as ab 106582 (abcam, Cambridge, MA); anti-FcRn antibodies such as sc-271745 (Santa Cruz Biotechnology, Santa Cruz, CA); anti-transferrin receptor antibodies such as ab61021 (abcam, Cambridge, MA); anti-insulin receptor antibodies such as ab5500 (abcam, Cambridge, MA); anti-IGF-1 receptor antibodies such as ab5681 (abcam, Cambridge, MA); anti-leptin receptor antibodies such as ab5593 (abcam, Cambridge, MA); anti-TNF alpha antibodies such as ab31908 (abcam, Cambridge, MA); anti-amyloid beta antibodies such as ab2539 (abcam, Cambridge, MA); anti-hyaluronic acid antibodies such as ab53842 (abcam, Cambridge, MA); anti-BACE antibodies such as ab2077 (abcam, Cambridge, MA); anti-TSG-6 antibodies such as ab204049 (abcam, Cambridge, MA).

[0152] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0153] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0154] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment.

In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0155] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0156] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0157] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0158] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0159] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0160] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody

binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-1 and antagonizes a PD-1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0161] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0162] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0163] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0164] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0165] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0166] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0167] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0168] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0169] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to PD-L1 and antagonizes a PD-L1-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0170] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0171] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0172] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and

antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0173] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0174] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0175] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0176] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0177] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0178] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety

on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TIM3 and antagonizes a TIM3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0179] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0180] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0181] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0182] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0183] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0184] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain

that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0185] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0186] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3 -mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0187] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to LAG3 and antagonizes a LAG3-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0188] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0189] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0190] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM,

IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0191] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0192] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0193] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0194] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0195] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0196] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD137 and agonizes a CD137-mediated T-cell inhibitory signaling

pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0197] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0198] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0199] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0200] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0201] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0202] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety

on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0203] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0204] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0205] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to OX40 and agonizes an OX40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0206] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0207] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0208] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0209] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0210] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0211] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0212] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0213] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0214] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD40 and agonizes a CD40-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0215] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR

and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0216] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0217] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0218] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0219] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0220] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has

an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0221] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0222] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0223] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GITR and agonizes a GITR-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0224] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0225] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0226] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0227] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that

comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0228] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0229] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0230] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0231] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0232] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD27 and agonizes a CD27-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0233] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or

IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment.

[0234] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0235] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0236] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0237] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0238] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes

an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0239] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0240] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0241] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HVEM and agonizes an HVEM-mediated T-cell inhibitory signaling pathway has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0242] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EGFR has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0243] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER2 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0244] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises a human

serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to HER3 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0245] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to EPCAM has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0246] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CEACAM has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0247] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP100 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to GP 100 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0248] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to MAGE1 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0249] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to NY-ESO-1 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0250] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Sialyl Lewis X antigen has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0251] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to Tn antigen has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0252] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD19 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0253] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises

a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD20 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0254] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD22 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0255] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD33 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0256] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD38 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0257] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In

one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD52 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0258] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to CD70 has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0259] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises an Fc domain. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid binding protein (HABP). In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises TSG-6. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a TSG-6-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to VEGF has an ADME-modulating moiety on the J-chain that comprises a TSG-6-binding scFv antibody fragment.

[0260] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises an Fc domain. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid binding protein (HABP). In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises TSG-6. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has

an ADME-modulating moiety on the J-chain that comprises a hyaluronic acid-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a TSG-6-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to TNF alpha has an ADME-modulating moiety on the J-chain that comprises a TSG-6-binding scFv antibody fragment.

[0261] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0262] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0263] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0264] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0265] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0266] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0267] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0268] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to amyloid beta has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0269] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises human serum albumin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding peptide. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a human serum albumin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an FcRn-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an Fc domain.

[0270] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises transferrin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a transferrin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a transferrin receptor-binding scFv antibody fragment.

[0271] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises leptin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a leptin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a leptin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a leptin receptor-binding scFv antibody fragment.

[0272] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises insulin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an insulin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an insulin-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE

has an ADME-modulating moiety on the J-chain that comprises an insulin receptor-binding scFv antibody fragment.

[0273] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises IGF-1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an IGF-1-binding scFv antibody fragment. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises an IGF-1 receptor-binding scFv antibody fragment.

[0274] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises basigin. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a basigin-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a basigin-binding scFv antibody fragment.

[0275] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises Glut1. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a Glut1-binding scFv antibody fragment.

[0276] In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises CD98hc. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding antibody moiety. In one specific embodiment, a binding molecule whose IgM, IgA, IgG/IgM, or IgG/IgA antibody binds to BACE has an ADME-modulating moiety on the J-chain that comprises a CD98hc-binding scFv antibody fragment.

[0277] It is to be understood that an IgM, IgA, IgG/IgM or IgG/IgA antibody that binds to any of the listed targets described herein can be combined with a modified J-chain with any of the ADME-modulating moieties listed herein to create a binding molecule. Thus, any antibody target listed herein can be combined with any ADME-modulating moiety listed herein. FIG. 13 provides a list of non-limiting examples of antibody targets and ADME-modulating moieties that can be included on a J-chain of a binding molecule in accordance with aspects of the invention. Any of the antibody targets listed in the left column of FIG. 13 can be combined with any of the ADME-modulating moieties listed in the right column of FIG. 13.

[0278] While certain preferred embodiments are specifically referred to herein, it is to be understood that IgM, IgA, IgG/IgM and IgG/IgA antibodies with binding specificity to any target, such as any tumor antigen, comprising a modified J-chain with any ADME-modulating moiety described herein are contemplated and are within the scope of the present invention.

[0279] In a preferred embodiment, the multi-specific IgM, IgA, IgG/IgM or IgG/IgA antibody binds to one or more of the tumor targets listed herein, while the J-chain comprises an ADME-modulating moiety.

[0280] In another preferred embodiment, the J-chain of the subject binding molecules includes an ADME-modulating moiety that is an scFv, and that reduces clearance of the binding molecule by binding to albumin. In one preferred embodiment, the ADME-modulating moiety on the J-chain is an scFv that binds to albumin.

[0281] In one preferred embodiment, a binding molecule includes an IgM antibody that binds to CD20, and the ADME-modulating moiety on the J-chain is human serum albumin (HSA). In another preferred embodiment, a binding molecule includes an IgM antibody that binds to CD20, and the ADME-modulating moiety on the J-chain is an anti-albumin scFv.

[0282] In one preferred embodiment, a binding molecule includes an IgM antibody that binds to DR5, and the ADME-modulating moiety on the J-chain is human serum albumin (HSA). In another preferred embodiment, a binding molecule includes an IgM antibody that binds to DR5, and the ADME-modulating moiety on the J-chain is an anti-albumin scFv.

[0283] In one preferred embodiment, a binding molecule includes an IgM antibody that binds to BACE, and the ADME-modulating moiety on the J-chain is transferrin. In another preferred embodiment, a binding molecule includes an IgM antibody that binds to BACE, and the ADME-modulating moiety on the J-chain is an anti-transferrin receptor scFv. In one preferred embodiment, a binding molecule includes an IgM antibody that binds to BACE, and the ADME-modulating moiety on the J-chain is an anti-transferrin scFv.

[0284] In one preferred embodiment, a binding molecule includes an IgM antibody that binds to VEGF, and the ADME-modulating moiety on the J-chain is hyaluronic acid binding protein (HABP). In another preferred embodiment, a binding molecule includes an IgM antibody that binds to VEGF, and the ADME-modulating moiety on the J-chain is an anti-

hyaluronic acid scFv.

[0285] In one preferred embodiment, a binding molecule includes an IgM antibody that binds to TNF alpha, and the ADME-modulating moiety on the J-chain is hyaluronic acid binding protein (HABP). In another preferred embodiment, a binding molecule includes an IgM antibody that binds to TNF alpha, and the ADME-modulating moiety on the J-chain is an anti-hyaluronic acid scFv.

[0286] In all embodiments, an ADME-modulating moiety of the modified J-chain may be introduced before or after the J-chain. Thus, a modified J-chain with an anti-albumin scFv ADME-modulating moiety that increases the retention of the binding molecule in the circulation by binding to albumin may have an anti-albumin scFv-J or a J-anti-albumin scFv configuration. A schematic illustration of two non-limiting examples of such configurations are provided in FIGS. 4A and 4B.

[0287] Due to their increased avidity, the subject binding molecules are superior relative to bispecific IgG antibodies. For example, as a result, they are suitable for targeting low level expression targets, such as Rituxan-resistant Burkitt lymphoma cells characterized by a low level of CD20 expression. In addition, the IgM, IgA, IgG/IgM and IgG/IgA antibodies herein comprising a modified J-chain have greatly enhanced potency relative to bispecific IgG antibodies.

Pharmaceutical Compositions of Antibodies with Modified J-Chain

[0288] For therapeutic uses, the subject binding molecules can be formulated into pharmaceutical compositions. A pharmaceutical composition of the present invention can be administered by a variety of methods known in the art. As will be appreciated by the skilled artisan, the route and/or mode of administration will vary depending upon the target disease or condition and the desired results. To administer a compound of the invention by certain routes of administration, it may be necessary to coat the compound with, or co-administer the compound with, a material to prevent its inactivation. For example, the compound may be administered to a subject in an appropriate carrier, for example, liposomes, or a diluent. Pharmaceutically acceptable diluents include saline and aqueous buffer solutions. Pharmaceutical carriers include sterile aqueous solutions or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersion. The use of such media and agents for pharmaceutically active substances is known in the art.

[0289] The compositions may also contain adjuvants such as preservatives, wetting agents, emulsifying agents and/or dispersing agents. Prevention of presence of microorganisms may be ensured both by sterilization procedures and by the inclusion of various antibacterial and antifungal agents, for example, paraben, chlorobutanol, phenol, sorbic acid, and the like. It may also be desirable to include isotonic agents, such as sugars, sodium chloride, and the like into the compositions. In addition, prolonged absorption of the injectable pharmaceutical form may be brought about by the inclusion of agents which delay absorption such as aluminum monostearate and gelatin.

[0290] Actual dosage levels of the active ingredients in the pharmaceutical compositions of the present invention may be varied so as to obtain an amount of the active ingredient which is effective to achieve the desired therapeutic response for a particular patient, composition, and mode of administration, without being toxic to the patient. The selected dosage level will depend upon a variety of pharmacokinetic factors including the activity of the particular compositions of the present invention employed, the route of administration, the time of administration, the rate of excretion of the particular compound being employed, the duration of the treatment, other drugs, compounds and/or materials used in combination with the particular compositions employed, the age, sex, weight, condition, general health and prior medical history of the patient being treated, and like factors well known in the medical arts.

[0291] The composition must be sterile and fluid to the extent that the composition is deliverable by syringe. In addition to water, the carrier preferably is an isotonic buffered saline solution.

[0292] The following examples, sequence listing and figures are provided to aid the understanding of the present invention, the true scope of which is set forth in the appended claims.

[0293] Further details of the invention are illustrated by the following non-limiting Examples.

Example 1: IgMs can be conjugated to multiple scFvs on either end of the J-chain with no effect on functional activity

[0294] The J-chain of an IgM molecule can be linked in frame with an scFv designed to bind a target of interest at either its C- or N-terminus, and the resultant bispecific IgMs are not perturbed in structure or function as evidenced by no diminution in their CDC activity.

1. Generation of DNA constructs with designed mutations

[0295] *DNA construct synthesis.* All the DNA constructs with designed mutations are synthesized by commercial vendors (Genescript), with compatible restriction sites at both ends for sub-cloning into respective expression vectors.

[0296] *Constructing expression vectors.* The synthesized DNA constructs are re-suspended in Tris-EDTA buffer at 1

μg/ml. DNA (1 μg) is subjected to enzyme digestion and the synthesized gene is separated from the carrier plasmid DNA by electrophoresis. The digested DNA is ligated to pre-digested plasmid DNA (pCAGGS for J-chain, Gene 108 (1991) 193-200) by standard molecular biology techniques. The ligated DNA is transformed into competent bacteria and plated on LB plates with multiple selective antibiotics. Several bacterial colonies are picked and DNA preparations are made by standard molecular biology techniques. The prepared DNA are verified by sequencing. Only the bacterial clones with 100% match of DNA sequence with the designed DNA sequence are used for plasmid DNA preparation and subsequently for cell transfection.

[0297] IgM heavy chain: This heavy chain construct has a full length μ chain for an anti-CD20 IgM which binds CD20 on the surface of B-cells:

IgM Heavy chain sequence of an anti-CD20 antibody:

MGWSYIILFLVATATGVHSQVQLQQPGAELVKPGASVKMSCKASGYTFTSYNMHWV
KQTPGRGLEWIGAIYPGNGDTSYNQKFKGKATLTADKSSSTAYMQLSSLTSEDSAVY
YCARSTYYGGDWYFNVWGAGTTVTVSSGSASAPTLFPLVSCENSPSDTSSVAVGCLA
QDFLPDSITFSWKYKNNSDISSTRGFPSVLRGGKYAATSQVLLPSKDVMQGTDEHVV
CKVQHPNGNKEKNVPLPVIAELPPKVSVFVPPRDGFFGNPRKSKLICQATGFSPRQIQV
SWLREGKQVGSGVTTDQVQAEAKESGPTTYKVTSTLTIKESDWLSQSMFTCRVDHR
GLTFQQNASSMCVPDQDTAIRVFAIPPSFASIFLTKSTKLTCLVTDLTTYDSVTISWTR
QNGEAVKTHTNISESHPNATFSAVGEASICEDDWNSGERFTCTVTHTDLPSPLKQTISR
PKGVALHRPDVYLLPPAREQLNLRESATITCLVTGFSPADVFVQWMQRGQPLSPEKY
VTSAPMPEPQAPGRYFAHSILTVSEEEWNTGETYTCVVAHEALPNRVTERTVDKSTG
KPTLYNVSLVMSDTAGTCY (SEQ ID: 16)

[0298] This heavy chain construct has a molecular weight about 64 kD and when co-expressed with light chain, the resultant IgM is able to bind to CDIM positive B cells.

IgM Light chain sequence of an anti-CD20 antibody:

MDMRVPAQLLGLLLLWLRGARCQIVLSQSPAILSASPGEKVTMTCRASSSVSYIHWF
QQKPGSSPKPWIYATSNLASGVPVRFSGSGSGTSYSLTISRVEAEDAATYYCQQWTSN
PPTFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDN
ALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFN
RGEC (SEQ ID NO: 17)

[0299] The light chain construct has a molecular weight of about 24kD and when co-expressed with the appropriate heavy chain (SEQ ID NO: 16) is able to bind to CDIM positive B cells.

[0300] *Different J-chains.* In order to demonstrate that J-chain variants were able to couple with IgM, two different J-chain variants are constructed with distinct fusion sites incorporating anti-CD3 antibody (OKT3 scFv).

i. This construct is composed of an scFv of OKT3 (anti-CD3) fused with N-terminus of human J-chain (CD3scFv-15 aa Linker-J, O15J):

QVQLQQSGAELARPGASVKMSCKASGYTFTRYTMHWVKQRPGQGLEWIGYINPSRG
YTNYNQKFKDKATLTTDKSSSTAYMQLSSLTSEDSAVYYCARYYDDHYSLDYWGQ
GTTLTVSSGGGGSGGGGSGGGGSQIVLTQSPAIMSASPGEKVTMTCSASSSVSYMNW
YQQKSGTSPKRWIYDTSKLASGVPAHFRGSGSGTSYSLTISGMEAEDAATYYCQQWS
SNPFTFGSGTKLEIKGGGGSGGGGSGGGGSQEDERIVLVDNKCKCARITSRIIRSSEDP
NEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQS
NICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPDGGGSEQKLI
SEEDLNSAVDHHHHHH (SEQ ID NO: 18)

[0301] This construct has a molecular weight about 45kD and is able to bind to soluble epsilon chain of CD3 (Sino Biological), or T cells; and is able to bind to anti-myc monoclonal antibody 4A6 or other anti-myc antibodies.

[0302] ii. This construct is composed of a scFv of OKT3 (anti-CD3) fused with C-terminus of human J-chain (J-15 aa Linker-CD3scFv, J15O):

QEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFV
YHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYG
GETKMVETALTPDACYPDGGGGSGGGGSGGGGSQVQLQQSGAELARPGASVKMSC
KASGYTFTRYTMHWVKQRPGQGLEWIGYINPSRGYTNYNQKFKDKATLTTDKSSST

AYMQLSSLTSEDSAVYYCARYYDDHYSLDYWGQGTTLTVSSGGGGSGGGGSGGGG
SQIVLTQSPAIMSASPGEKVTMTCSASSSVSYMNWYQQKSGTSPKRWIYDTSKLASG
VPAHFRGSGSGTSYSLTISGMEAEDAATYYCQQWSSNPFTFGSGTKLEIKEQKLISEED
LNSAVDHHHHHH- (SEQ ID NO: 19)

[0303] This J-CD3scFv construct has a molecular weight about 45kD and is able to bind to soluble epsilon chain of CD3 (Sino Biological), or T cells; and is able to bind to anti-myc monoclonal antibody 4A6 or other anti-myc antibodies.

[0304] To establish that assembly of bispecific IgM is feasible with a modified J-chain carrying an anti-CD3 scFv of a different sequence than that used in Examples 1 and 2, a J-chain carrying the variable regions from the antibody Visilizumab (Nuvion) was performed. Shown below are the sequences for two J-chains with the scFv corresponding to Visilizumab (V) fused to the J-chain through a linker containing 15 amino acid residues in two different orientations - V15J and J15V.

[0305] J chain sequence for V15J:

MGWSYIILFLVATATGVHSQVQLVQSGAEVKKPGASVKVSCKASGYTFISYTMHWV
RQAPGQGLEWMGYINPRSGYTHYNQKLKDKATLTADKSASTAYMELSSLRSEDTAV
YYCARSAYYDYDGFAYWGQGTLVTVSSGGGGSGGGGSGGGGSDIQMTQSPSSLSAS
VGDRVTITCSASSSVSYMNWYQQKPGKAPKRLIYDTSKLASGVPSRFSGSGSGTDFTL
TISSLQPEDFATYYCQQWSSNPPTFGGGTKLEIKGGGGSGGGGSGGGGSQEDERIVLV
DNKCKCARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCK
KCDPTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVET
ALTPDACYPD (SEQ ID NO: 20)

[0306] J-chain sequence for J15V:

MKNHLLFWGVLAVFIKAVHVKAQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVER
NIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDEDS
ATETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPDGGGGSGGGGSGGGG

SQVQLVQSGAEVKKPGASVKVSCKASGYTFISYTMHWVRQAPGQGLEWMGYINPRS
GYTHYNQKLKDKATLTADKSASTAYMELSSLRSEDTAVYYCARSAYYDYDGFAYW
GQGTLVTVSSGGGGSGGGGSGGGGSDIQMTQSPSSLSASVGDRVTITCSASSSVSYMN
WYQQKPGKAPKRLIYDTSKLASGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQWS
SNPPTFGGGTKLEIK (SEQ ID NO: 21)

[0307] DNA corresponding to these sequences was synthesized and transfected into HEK293 cells along with the heavy and light chains for anti-CD20 IgM to produce protein which was then purified using the camelid antibody affinity matrix specific for IgM. As shown in FIG. 6, J-chains fused to the new anti-CD3 scFv with the 15 aa linker are able to incorporate into the IgM and the pentameric form of bi-specific IgM with the corresponding J-chain is clearly distinguishable from the hexameric form without a J-chain.

[0308] 2. Protein expression, purification and characterization

a. Transfection. Heavy, Light and Modified J-chain DNA is transfected into CHO cells. DNA for expression vectors are mixed typically in 1:1:1 ratio with PEI and then added to CHO-S cells. PEI transfection with CHO-S cells is conducted according to established techniques (see Biotechnology and Bioengineering, Vol 87, 553-545).

b. Immunoprecipitation

i. Capture Select IgM (BAC, Thermo Fisher). IgM proteins from transfected CHO cell supernatants are partially purified by immunoprecipitation with Capture Select IgM affinity matrix according to manufacturers' protocol (GE Life Sciences). After incubation at room temperature for 2 hours, the affinity matrix is separated from the supernatant by centrifugation. The matrix is further washed with PBS for 3 times before the PBS is carefully removed. The captured protein is eluted from the matrix by incubating with NuPage LDS protein buffer (Life Technology) for 5 minutes.

ii. Anti-myc agarose affinity matrix (Sigma). IgM proteins from transfected CHO cell supernatants are partially purified by immunoprecipitation with anti-myc affinity matrix according to manufacturers' protocol. After incubation at room temperature for 2 hours, the affinity matrix is separated from the supernatant by centrifugation. The matrix is further washed with PBS for 3 times before the PBS is carefully removed after the final wash. The captured protein is eluted from the matrix by incubating with NuPage LDS protein buffer (Life Technology) for 5 minutes.

c. Gel electrophoresis

i. Non-reducing SDS PAGE separates native IgM and its mutant forms according to size. Pentameric IgM, composed of homodimeric heavy and light chains, produces a protein band of approximately 1,000,000 molecular weight. NuPage LDS Sample Buffer (Life Technologies) is added to IgM protein samples at 25 C for 30 minutes before loading onto the gel. NativePage Novex 3-12% Bis-Tris Gel (Life Technologies) is used with Novex Tris-Acetate SDS Running Buffer (Life Technologies). Run gel until the dye front reaches the bottom of the gel.

ii. Reducing SDS-PAGE. NuPage LDS sample buffer (Life Technologies) and NuPage reducing agent dithiothreitol (Life Technologies) are added to IgM protein samples and heated to 80°C for 10 minutes before loading on NuPage Novex 4-12% Bis-Tris Gel (Life Technologies). NuPage MES SDS Running Buffer (Life Technologies) is used for gel electrophoresis. Gels are run until the dye front reaches the bottom of the gel. After electrophoresis is complete, remove gel from apparatus and stain the gel using Colloidal Blue Staining (Life Technologies).

iii. The DNA corresponding to these heavy and light chains as well as that corresponding to either the wild-type (wt) J-chain, O15J or 1150 J-chain sequences described above were co-transfected into HEK293 cells and proteins expressed and purified using the camelid resin as described before. As shown in FIG. 6, all four proteins express well. The anti-CD20 IgM hexamer without J-chain is clearly resolved from the J-chain containing pentamers for the IgM pentamer with the wild type J-chain as well as for the bispecific IgMs where the anti-CD3 scFv is linked to the J-chain in either orientation (O15J or 1150).

Analysis of complement dependent cytotoxicity for family of IgMs with and without incorporated J-chains

[0309] Complement dependent cytotoxicity is a key mechanism for cell killing by antibodies. IgM antibodies are known to have enhanced complement dependent cell killing (CDC) due to their multimeric form. A key aspect of this invention was to test if incorporation of modified J-chains, which carry scFv or camelid Vhh binders of effector cells at either their C- or N- termini, causes interference with binding of C1q - the key component of the complement pathway, and therefore may inhibit CDC. The CDC activity of each of the IgM and bispecific IgM constructs was measured. As shown in FIG. 7, incorporation of the modified J-chain has, unexpectedly, no deleterious effect on the CDC activity of the bispecific IgMs. Moreover, with the linker lengths tested, it was found that the bispecific IgMs have CDC activity between 60-100 fold enhanced over the corresponding IgG on a molar basis (FIG. 7).

Example 2: Bispecific IgMs can bind two targets simultaneously and show functional effects

[0310] The DNA corresponding to these heavy and light chains as well as that corresponding to either the wild-type (wt) J-chain (FIG. 3), V15J or J15V J-chain sequences shown above were co-transfected into HEK293 cells and proteins expressed and purified using the camelid resin as described before. As shown in FIG. 6, all four proteins express well. The anti-CD20 IgM hexamer without J-chain is clearly resolved from the J-chain containing pentamers for the IgM pentamer with the wild type J-chain as well as for the bispecific IgMs where the anti-CD3 scFv is linked to the J-chain in either orientation.

[0311] Purified proteins were analyzed for T-cell activation using a commercially available Luciferase reporter gene based kit (Promega). Briefly, purified protein was added to 7500 Ramos and 25000 engineered Jurkat cells (Promega CS176403) in 40uL RPMI with 10% FBS. Mixture was incubated for 5h 37C with 5% CO_2. Cells were mixed with lysis buffer containing luciferin to measure luciferase reporter activity. Light output was measured by EnVision plate reader and analyzed by Prism software. As shown in FIG. 8, only the antibodies that carried the CD3 specific scFv binding moiety on the J-chain are able to show dose dependent activation, whereas the IgM antibody lacking the modified J-chain or the IgG are unable to show any signal in this assay.

Example 3: Construction and testing of anti-CD20 antibody with albumin binding domain tethered to J-chain

[0312] The half-life of IgMs in human plasma is estimated to be around 2-3 days and shorter still in mice (FIG. 9). This is significantly shorter than for IgGs, which interact with the neonatal Fc receptor (FcRn) and are recycled after endocytosis enabling a much longer half-life of roughly 21 days. In order to increase the half-life of IgMs, tethering of scFvs to either terminus of the J-chain was performed, without significantly altering the effector functions of IgMs such as CDC (FIG. 7).

[0313] There are several approaches that have been described in the art to enable half-life extension of biologics. These include tethering of mutants of human serum albumin (Andersen et al, JBC VOL. 289, NO. 19, pp. 13492-13502, 2014), peptides (Dennis et al, J. Biol. Chem. 2002, 277:35035-35043) or scFvs that can bind human serum albumin (Muller et al mAbs 4:6, 673-685; 2012),

[0314] Shown below is the sequence of an example J-chain that can be used to extend the half-life of IgMs by utilizing

an albumin binding domain designed for binding to human serum albumin with high affinity (Hopp et al PEDS 23:pp 827-833 (2010)).

[0315] Albumin binding domain: QHDEAVDANSLAEAKVLANRELDKYGVSDYYKNLINNAKTVEGVKALIDEILAALP (SEQ ID NO: 22)

[0316] Wt J-chain:

QEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFV YHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYG GETKMVETALTPDACYPD (SEQ ID NO: 1)

[0317] A15J:

QHDEAVDANSLAEAKVLANRELDKYGVSDYYKNLINNAKTVEGVKALIDEILAALP GGGGSGGGGSGGGGSQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIRIIVPLN NRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSATETCYTY DRNKCYTAVVPLVYGGETKMVETALTPDACYPD (SEQ ID NO: 23)

[0318] Expression and assembly of this ABD-J-chain fusion into IgMs was tested using the IgM sequence described in Example 1. In addition, fusion of this ABD to J-chain was verified not to perturb the CDC activity on anti-CD20 IgM on target cell lines carrying CD20 on their surface (eg. Ramos) as described in Example 1. Finally, the affinity of ABD in the context of the IgM, for binding to HSA was measured using immobilized HSA using surface plasmon resonance (Biacore).

Example 4: Construction and testing of anti-CD20 antibody with transferrin binding scFv

[0319] Delivery of biologic drugs to targets in the central nervous system, particularly the brain, is a challenging problem because of the Blood Brain Barrier (BBB). The transferrin receptor (TfR) is overexpressed in the endothelium of the BBB. It is thought to act as a shuttle to transport nutrients such as iron from the periphery to the brain. Receptor mediated transcytosis (RMT) has been used by several groups to deliver biologics to the brain. For example, Jones et al have described the use of transferrin binding antibodies as a method of shuttling biologics across the BBB (Jones, A.R., and E.V. Shusta. 2007. Blood-brain barrier transport of therapeutics via receptor-mediation. Pharm. Res. 24:1759-1771).

[0320] One such transferrin binding sequence was used (Vh sequence selected from phage display by Yang et al) to make an in-frame fusion with our J-chain as shown below.

[0321] Transferrin receptor binding Vh sequence:

MAQVQLLESGGGLVQPGGSLRLSCAASGFIFNTEYMAWVRQ APGKGLEWVSAIKEQSGSTYYADSVKGRFTISRDNSKNTLYL QMNSLRAEDTAVYYCAAQMHHEAEVKFWGQGTLVTVS (SEQ ID NO: 24)

[0322] Transferrin receptor binding Vh sequence fused to J-chain at N-terminus:

MAQVQLLESGGGLVQPGGSLRLSCAASGFIFNTEYMAWVRQ
APGKGLEWVSAIKEQSGSTYYADSVKGRFTISRDNSKNTLYL
QMNSLRAEDTAVYYCAAQMHHEAEVKFWGQGTL
VTVSGGGGSGGGGSGGGGSQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIRII
VPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSATET
CYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPD (SEQ ID NO: 25)

[0323] The fusion J-chain was incorporated into a relevant IgM (for example the CD20 IgM described previously). In addition to the assays described earlier for expression and assembly, antigen binding, cell binding and cell internalization assays were carried out to verify that the resultant IgM+TfR J-chain is functional.

[0324] Antigen binding was tested using ELISAs with commercially available recombinant human transferin receptor (R&D Systems) immobilized on plates. Briefly, ~100 ng of human tansferrin receptor was added to a 96-Wellplate (Nunc Maxisorb plate) per well at 4C, overnight. The plate was washed with PBS-0.05% Tween-20 three times and blocked with StartingBlock (Pierce) at 37C for 1 hour. Then the plate was washed with PBST three times after the blocking solution was removed. The bispecific antibodies with different concentrations were added to each well and the plate was allowed to stand at 37C for 1 hour. After three PBST washes, HRP-conjugated anti-human IgG Fc antibody (Abcam, diluted in StartingBlock at a ratio of 1:10,000) was added to each well, and the plate was further incubated at 37° C for 1 hour. After three PBST washes, colorimetric TMB substrate (US Biological) was added to each well to perform a peroxidase reaction. After the addition of stop solution (1 M H2SO4), the absorbance was monitored at 450 nm and the equilibrium constant (K_D) for the antibody was calculated by fitting the resultant data with Graph Pad Prism. For testing CD20 binding, an ELISA using immobilized CD20-Fc (Acros Biosystems) was used as illustrated in FIG. 10. Detection antibody for this ELISA is a mouse anti-human kappa light chain antibody conjugated with HRP (Southern Biotech, 9230-05). Capture, detection and development are carried out as detailed above.

[0325] To verify that the resultant IgM binds to target cells by using FACS based assay such as those described in Example 1, on tumor cell lines known to overexpress the transferrin receptor for example the human erythroleukemia cell line K562. Mean fluorescence intensity readings were analyzed using GraphPad Prism to calculate a K_d.

Example 5: Use of site specific chemoenzymatic labeling to generate imaging agents and antibody drug conjugates with IgMs

[0326] IgMs are very large biomolecules (>1 MDa with J-chain). Labeling of IgMs to enable visualization in animal studies is problematic because of the numerous free lysine residues. In order to enable labeling with stoichiometry and positions that retain the activity of IgMs, site specific labeling is carried out using chemo-enzymatic approaches as reviewed in Kline et al (Pharm Res 2014 Dec 16).

[0327] One method for site specifically labeling IgM molecules is to use a glycan labeling strategy as described in Houghton et al (PNAS (52) 15850-15855). The method uses a combination of enzymes - beta galactosidase to remove a terminal galactose residue and then a promiscuous galactose transferase (GalTY289L) to install an azide labeled sugar (GlcNAz) that can be used to post-synthetically add a DIBO labeled dye or cytotoxin. Because the heavy chain of IgMs carries five glycans as opposed to the single glycan on each heavy chain of an IgG antibody, much more efficient labeling is expected using this approach with an antibody to dye/drug ratio of up to 1:102 if the glycan on the J-chain is also derivatized. As shown in FIG. 10, using an example IgM (1.5.3V15J15HSA) efficient labeling was demonstrated with this approach, and an Alexa 647 DIBO dye. Clearly, a similar approach can also be used to generate IgMs that are labeled with PET tracers and cytotoxic molecules.

[0328] As a second example of using an acceptor sequence on the J-chain for post-translational site-specific labeling, the "LLQGA" recognition site of microbial transglutaminase (mTGase) is added to the C-terminus of J-chain as shown below (FIG.12).

[0329] J chain with "Q Tag":

QEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFV
YHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYG
GETKMVETALTPDACYPDGGGGSGGGGSGGGGSLLQGA (SEQ ID NO: 26)

[0330] Next, dye molecules with a primary amine at its terminus e.g Alexa 488 Cadaverine (Thermo Scientific) was reacted with IgMs incorporating this J-chain in the presence of mTGAse under standard conditions (Strop et al Bioconjugate Chemistry 2015 26(4) 650-9). After incubation overnight at room temperature with 5X molar excess of dye, free dye was separated from labeled IgM using size exclusion chromatography on a NAP-5 column (Biorad). Incorporation of dye was quantitated using absorbance at 488 nm.

[0331] It follows that such methods can also be used with other enzymes that can be used for chemo-enzymatic modification as well as other small molecules (e.g., cytotoxic drugs) that carry appropriate handles for functionalization.

Example 6: In vivo bio-distribution studies using IgMs labeled with near infra-red dye VivoTag680 (Perkin Elmer)

[0332] In order to follow the bio-distribution of IGM-55.5 in mice, the molecule was labeled with a near infra-red dye VivoTag680 (Perkin Elmer) using standard amine coupling with an NHS ester at neutral pH (Vasquez et al, PLoS One. 2011; 6: e20594). The injected group received an intravenous injection with 2 nmol/mouse of the labeled IgM molecule. The background control group remained un-injected as a way to distinguish the fluorescence signal of the labeled antibodies from low level background signal, which is primarily from food in the gut. The t0 imaging time point was performed immediately after injection with antibody. Mice were sacrificed after the final in vivo imaging timepoint, followed by resection of tissues and ex vivo imaging.

[0333] A generalized schematic of a temporal biodistribution model assessed by in vivo 3D FMT is presented in FIG. 11 Panel A. This type of study is well suited to non-invasively determine both the blood PK of labeled antibodies (determined from the decrease in fluorescence signal of blood in the heart), as well as kinetic biodistribution into various organ systems (brain, lungs, heart, liver, kidneys, stomach, intestines, bladder, and skin). For each animal at each time point, the blood fluorescence signal was subtracted from the total signal of each of the other organs to provide a more accurate determination of tissue accumulation. The in vivo tissues were also assessed ex vivo at the terminal time point by epifluorescence. Ex vivo epifluorescence measurements were also obtained for gall bladder, muscle, spleen, pancreas, white blood cells, lymph nodes, and intestines (which were flushed prior to imaging to remove fecal material).

[0334] Whole body and head bio-distribution imaging was performed on the FMT4000 at 0, 1, 2, 4, 8, 24, 48, and 96 h post-injection. Additional animals were bled at 0, 1, 2, 4, 8, 24, 48, 96h, and these blood samples were shipped to IGM Biosciences for assay. For tomographic imaging, animals were positioned in the supine position within an imaging cassette that provided gentle restraint and mild compression. All images were successfully acquired at the planned timepoints. Whole body non-invasive biodistribution and blood pharmacokinetics showed rapid blood clearance (t1/2 = 20 minutes) and dominant liver accumulation with some stomach and kidney signal. Un-injected controls showed only low level signal within the stomach and intestines, and data from IgM-injected mice were corrected for these background levels. The accumulation in liver, kidney and stomach was very rapid and achieved the highest levels at 1 h post-injection, partially clearing by 96 h. The majority of the signal resided in the liver (approximately 5X that of the other tissues); but when normalizing for tissue weight, comparable signal intensity could be seen in the stomach, with somewhat lower signal intensity in the kidneys (FIG. 11, Panel B). Such in vivo studies can also be carried out with the IgMs carrying modified J-chains to assess the increase in half-life or tissue distribution.

Example 7: Pharmacokinetics of IgG v. IgM with J-chain

[0335] Pharmacokinetic (PK) studies were conducted in Balb/c mice to assess clearance of IgG and IgM antibodies, with and without an attached modified J-chain. 100 ug of each antibody was administered to the mice via intravenous infusion. Approximately 500 uL of blood was collected by terminal cardiac puncture at each timepoint, with 3 mice per timepoint, and 8 or 15 timepoints total. ELISA was used to measure the concentration of each antibody in the blood. Quality metrics were verified on all ELISAs, and PK parameters were derived using standard curve fitting techniques.

[0336] PK results from Rituximab, polyclonal IgM and IgM 55.5 are provided in FIG. 16. These results demonstrate that IgM half-life in mice is significantly shorter that IgG half-life, as evidenced by the fact Rituximab (IgG) had a longer half-life than either the polyclonal IgM of the IgM 55.5. In addition, the half-life of IgM 55.5, produced in CHO cells, was shorter than that of human polyclonal IgM.

[0337] Results from IgM 1.5.3 with and without J-chain are provided in FIG. 17. As shown, the half-life of IgM 1.5.3 with no J-chain (1.5.3 IgM) was comparable to the half-life of IgM 55.5. The addition of a wild-type J-chain reduced the half-life of IgM 1.5.3. Addition of a J-chain having the V-linker-J orientation (1.5.3. V15J) further reduced the half-life of the antibody. These results demonstrate that the addition of J-chain to an IgM antibody reduces the half-life of the antibody.

Example 8: Fusion of an albumin binding domain to the J-chain significantly reduces clearance of IgMs

[0338] As noted above, the pharmacokinetics of IgMs indicate rapid blood clearance. Experiments were performed to determine the serum half-life-extending effects of tethering an albumin binding domain (ABD) (SEQ ID NO: 22) to an

IgM J-chain. DNA corresponding to the IgM heavy and light chains as shown in Example 1, as well as that corresponding to either the V15J sequence of Example 1 (Visilizumab (V) fused to the J-chain through a linker containing 15 amino acid residues) or the A15J sequence of Example 3 (an albumin-binding domain fused to the J-chain through a linker containing 15 amino acid residues) were co-transfected into HEK293 cells, and the proteins were expressed and purified using the camelid resin as described before. Three groups of mice received an intravenous injection with 100 ug/mouse of either V15J-1.5.3-IgM, A15J-1.5.3-IgM, or Rituximab (IgG). Blood samples were taken periodically following the initial injection, and the serum concentration of each injected antibody was measured in the samples using an ELISA that was adapted to measure the concentration of the tested antibodies in serum.

[0339] The data demonstrate that fusion of an albumin-binding domain to the J-chain resulted in a significant and relatively large increase in the half-life of IgMs. As shown in FIG. 18, the beta half-life of V15J-1.5.3-IgM, which did not include the albumin binding domain, was only 7 hours. By contrast, the beta half-life of A15J-1.5.3-IgM, which did include the albumin-binding domain on the J-chain, was 32 hours, which was comparable to Rituximab.

Example 9: IgM albumin J-chain assembly and expression

[0340] J-chain constructs that incorporate a human serum albumin (HSA) were prepared as provided in Example 1. Constructs were prepared with the HSA positioned at the N-terminus of the J-chain (HSA-15-J), and at the C-terminus of the J-chain (J-15-HSA). To verify that IgM antibodies incorporating J-chains containing HSA in either of these configurations could be assembled and expressed, SDS-PAGE gels under reducing conditions and Western blots were conducted.

[0341] Reducing SDS-PAGE: NuPage LDS sample buffer (Life Technologies) and NuPage reducing agent dithiothreitol (Life Technologies) were added to IgM protein samples and heated to 80°C for 10 minutes before loading on NuPage Novex 4-12% Bis-Tris Gel (Life Technologies). NuPage MES SDS Running Buffer (Life Technologies) was used for gel electrophoresis. Gels were run until the dye front reached the bottom of the gel. After electrophoresis was complete, the gel was removed from the apparatus and stained using Colloidal Blue Staining (Life Technologies).

[0342] Western Blot: An acrylamide gel run under conditions described above was washed in a 20% ethanol solution for 10 minutes and then the protein was transferred to an iBlot PVDF membrane (Invitrogen) using the iBlot Dry Blotting System (Invitrogen) at 20V for 10 minutes. After transfer the PVDF membrane was blocked using 2% bovine serum albumin, 0.05% Tween 20 for at least 12 hours. A 1/500 dilution of Pierce J-chain antibody (ThermoFisher) was added to the membrane, incubated for 1 hour, and then a 1/5000 dilution of peroxidase-conjugated goat anti-rabbit IgG (Jackson ImmunoResearch) was added and allowed to incubate in darkness for 30 minutes. Finally, Super Signal West Pico Chemiluminescent Substrate (ThermoFisher) was added to the blot and the resulting signal was visualized using the ChemiDoc-It HR410 Imaging System (UVP) or by exposing the blot to X-ray film.

[0343] The results are provided in FIG. 19, and demonstrate that J-chains having either of these configurations can be successfully incorporated into IgM antibodies, and that the resulting IgM antibodies can be assembled and expressed by CHO cells.

Example 10: CDC activity of ABD/HSA-containing J-chains

[0344] Complement-dependent cytotoxicity (CDC) assays were conducted using IgM antibodies incorporating a J-chain having the HSA-15-J (HSA at the N-terminus of the J-chain, followed by a 15 amino acid linker sequence) or the J-15-HSA (HSA at the C-terminus of the J-chain, preceded by a 15 amino acid linker sequence) configuration.

[0345] Ramos, a CD20+ cell line, was seeded in 96 well half area white plates at 25,000 cells/well. The protein under evaluation and human complement (5% final, Quidel) were added to initiate the CDC analysis and the number of viable cells were measured using Cell Titer Glo and manufacturer's protocol. Luminescence was measured on an Envision multimode reader (Perkin Elmer) using 0.1 s integration time per well. The percentage of viable cells was calculated by normalizing the luminescence values (Relative luminescence units - RLU) versus wells with no added test compound. Data were analyzed using GraphPad Prism and a four parameter fit with top and bottom values fixed at 100 and 0% viability respectively.

[0346] The results are provided in FIG. 20. The results demonstrate that the assembled IgM+HSA J-chain antibodies are functionally active in CDC assays in both orientations.

Example 11: Pharmacokinetics of J-HSA and HSA-J constructs

[0347] PK studies, as described above, were conducted in mice to evaluate the PK characteristics of IgM antibodies incorporating a J-chain having the HSA-15-J or the J-15-HSA orientation. The results are provided in FIG. 21 and FIG. 22. The results demonstrate an orientation effect, wherein the HSA positioned at the N-terminus (HSA-15-J orientation) had diminished half-life in comparison to the J-15-HSA orientation (HSA located at the C-terminus).

Example 12: Assembly and expression of "bidentate" J-chain constructs

[0348] Assembly and expression studies were conducted as described above in Example 9 for constructs containing both a CD3-binding moiety (abbreviated as "V") and a half-life extending moiety (either an albumin-binding domain protein, abbreviated "ABD", or a human serum albumin protein, abbreviated as "HSA"). These constructs are referred to as "bidentate" constructs. A summary of all the constructs that were evaluated is provided below in Table 10.

[0349] Constructs were prepared with the half-life extending moiety (e.g., the "ABD" or the "HSA") positioned at the C-terminus of the J-chain, and the CD3-binding moiety (e.g., "V") positioned at the N-terminus. To verify that IgM antibodies incorporating J-chains having any of these configurations could be assembled and expressed, SDS-PAGE gels under reducing conditions and Western blots were conducted, as described above. The results are provided in FIG. 23, and demonstrate that J-chains having either of these configurations can be successfully incorporated into IgM molecules, and that the resulting IgM molecules can be assembled and expressed by CHO cells.

Example 13: CDC activity of bidentate J-chain constructs

[0350] CDC assays, as described above in Example 10, were conducted using IgM antibodies incorporating the bidentate J-chains described above in Example 12. The results are provided in FIG. 24, and in FIG. 25. The results demonstrate that the bidentate J-chains that were evaluated did not diminish the CDC activity of the IgM antibodies that were tested.

Example 14: Pharmacokinetics of bidentate J-chain constructs

[0351] PK studies, as described above, were conducted in mice to evaluate the PK characteristics of the IgM antibodies incorporating the bidentate J-chains described above in Example 12. The results are provided in FIG. 26 and FIG. 27. The results demonstrate that both the V-J-ABD and V-J-HSA bidentate J-chains exhibited good alpha and beta half-life, and that the overall $AUC_{0\text{-}inf}$ showed an approximately 60% increase as compared to 1.5.3 IgM J-15-HSA.

Example 15: In-vivo activity of bidentate J-chain constructs

[0352] CD34+ humanized NSG mouse studies were performed by In-Vivo Technologies, Inc. The mice were purchased from the Jackson Laboratory, and dosed with test articles through tail vein injection. Blood samples were collected at designated time points through facial vein. Blood samples from both the CD34+ mouse studies were sent back to IGM Biosciences Inc. for lymphocyte analysis. Blood samples were stained for human CD56, CD3, CD19 and CD45 markers to identify different populations of human lymphocytes. CountBright Absolute Counting Beads (LifeTechnologies, C36950) were used to quantify the absolute number of lymphocytes in the blood samples. The lymphocyte levels were plotted and analyzed using GraphPad Prism. As shown in FIG. 28, Panels A and B, the B-lymphocyte levels were essentially reduced to <10% of pre-dose levels, and this level was retained at the 24 hour timepoint for both 1.5.3V15J15HSA(K573P) and 1.5.3V15J15HSAwt with as little as 10 ug of article dosed one single time.

Table 10: Sequence Summary

SEQ ID NO:	Short Name	Sequence
27	Rituximab VH	QVQLQQPGAELVKPGASVKMSCKASGYTFTSYNMHWVKQTPGRGLEWIGAIYPGNGDT SYNQKFKGKATLTADKSSSTAYMQLSSLTSEDSAVYYCARSTYYGGDWYFNVWGAGTT VTVSA
28	Rituximab HCDR1	SYNMH
29	Rituximab HCDR2	AIYPGNGDTSYNQKFKG
30	Rituximab HCDR3	STYYGGDWYFNV
31	Rituximab VL	QIVLSQSPAILSASPGEKVTMTCRASSSVSYIHWFQQKPGSSPKPWIYATSNLASGVP VRFSGSGSGTSYSLTISRVEAEDAATYYCQQWTSNPPTFGGGTKLEIKR
32	Rituximab LCDR1	RASSSVSYIH
33	Rituximab LCDR2	ATSNLAS
34	Rituximab LCDR3	QQWTSNPPT
35	900 VH	EVQLVESGGG LVQPGGSLRL SCAASGYTFT SYNMHWVRQA PGKGLEWVGA IYPGNGDTSY NQKFKGRFTI SVDKSKNTLY LQMNSLRAED TAVYYCARVV YYSNSYWYFD VWGQGTLVTV SSASTKGPSV FPLAPSSKST SGGTAALGCL VKDYFPEPVT VSWNSGALTS GVHTFPAVLQ SSGLYSLSSV VTVPSSSLGT
36	900HCDR3	VVYYSNSYWYFDV

SEQ ID NO:	Short Name	Sequence
37	900 VL	DIQMTQSPSS LSASVGDRVT ITCRASSSVS YMHWYQQKPG KAPKPLIYAP SNLASGVPSR FSGSGSGTDF TLTISSLQPE DFATYYCQQW SFNPPTFGQG TKVEIKRTVA APSVFIFPPS DEQLKSGTAS VVCLLNNFYP REAKVQWKVD NALQSGNSQE SVTEQDSKDS TYSLSSTLTL SKADYEKHKV YACEVTHQGL
38	900LCDR1	RASSSVSYMH
39	900LCDR2	APSNLAS
40	900LCDR3	QQWSFNPPT
41	125 VH	EVQLVQSGAEVKKPGESLKISCKGSGRTFTSYNMHWVRQMPGKGLEWMGAIYPLTGDT SYNQKSKLQVTISADKSISTAYLQWSSLKASDTAMYYCARSTYVGGDWQFDVWGKGTT VTVSS
42	125HCDR2	AIYPLTGDTSYNQKSKL
43	125HCDR3	STYVGGDWQFDV
44	125 VL	EIVLTQSPGTLSLSPGERATLSCRASSSVPYIHWYQQKPGQAPRLLIYATSALASGIP DRFSGSGSGTDFTLTISRLEPEDFAVYYCQQWLSNPPTFGQGTKLEIK
45	125LCDR1	RASSSVPYIH
46	125LCDR2	ATSALAS
47	125LCDR3	QQWLSNPPT
48	844 VH #2	QVQLQQPGAELKKPGASVKVSCKASGYTFTSYNMHWVKQTPGRGLEWTGAIYPGNGDT SYNQKFKGKTTLTADKSSSTAYMELSSLRSEDTAVYYCARSTYYGGDWYFNVWGAGTT VTVSA
49	844 VH #3	QVQLQQPGAELKKPGASVKVSCKASGYTFTSYNMHWVKQTPGRGLEWIGAIYPGNGDT SYNQKFKGKTTLTADKSSSTAYMELSSLRSEDTAVYYCARSTYYGGDWYFNVWGAGTT VTVSA
50	844 VL #5	QIVLSQSPAIITASPGEKVTMTCRASTSASYIHWFQQKPTSSPKPWIYATSNLASGVP SRFSGSGSGTTYSMTISSLEAEDAATYYCQQWTSNPPTFGGGTKLEIK
51	844 VL #5 LCDR1	RASTSASYIH
52	844 VL #6	QIVLSQSPAIITASPGEKVTMTCRASTSVSYIHWFQQKPTSSPKPWIYATSNLASGVP SRFSGSGSGTTYSMTISSLEAEDAATYYCQQWTSNPPTFGGGTKLEIK
53	844 VL #6, #7 LCDR1	RASTSVSYIH
54	844 VL #7	QIVLSQSPAIITASPGEKVTMTCRASTSVSYIHWFQQKPGSSPKPWIYATSNLASGVP SRFSGSGSGTTYSMTISSLEAEDAATYYCQQWTSNPPTFGGGTKLEIK
55	844 VL #8	QIVLSQSPAIITASPGEKVTMTCRASSSVSYIHWFQQKPGSSPKPWIYATSNLASGVP SRFSGSGSGTTYSMTISSLEAEDAATYYCQQWTSNPPTFGGGTKLEIK
56	844 VH #10	EVQLQQSGAELKKPGASVKVSCKASGYTFTSYNMHWVKQTPGQGLEWIGAIYPGNGDT SYNQKFKGKTTLTADKSSSTAYMELSSLRSEDTAVYYCARSNYYGSSYWFFDVWGTGT TVTVSS
57	844 VH #10 HCDR3	SNYYGSSYWFFDV
58	844 VL #12	DIVLTQSPAIITASPGEKVTMTCRASSSVNYMDWYQKKPGSSPKPWIYATSNLASGVP SRFSGSGSGTTYSMTISSLEAEDAATYYCQQWSFNPPTFGGGTKLEIK
59	844 VL #12 LCDR1	RASSSVNYMD
60	844 VL #12 LCDR3	QQWSFNPPT
61	164 VH	QVQLQQSGAEVKKPGSSVKVSCKASGYTFTSYNMHWVKQAPGQGLEWIGAIYPGNGDT SYNQKFKGKATLTADESTNTAYMELSSLRSEDTAFYYCARSTYYGGDWYFDVWGQGTT VTVSS

SEQ ID NO:	Short Name	Sequence
62	164 VH HCDR3	STYYGGDWYFDV
63	164 VL	MGWSCIILFLVATATGVHSDIQLTQSPSSLSASVGDRVTMTCRASSSVSYIHWFQQKP GKAPKPWIYATSNLASGVPVRFSGSGSGTDYTFTISSLQPEDIATYYCQQWTSNPPTF GGGTKLEIK
64	1.5.3 VH	EVQLVQSGAEVKKPGESLKISCKGSGYSFTSYWIGWVRQMPGKGLEWMGIIYPGDSDT RYSPSFQGQVTISADKSITTAYLQWSSLKASDTAMYYCARHPSYGSGSPNFDYWGQGT LVTVSS
65	1.5.3 HCDR1	GYSFTSYWIG
66	1.5.3 HCDR2	IIYPGDSDTRYSPSFQG
67	1.5.3 HCDR3	HPSYGSGSPNFDY
68	1.5.3 VL	DIVMTQTPLSSPVTLGQPASISCRSSQSLVYSDGNTYLSWLQQRPGQPPRLLIYKISN RFSGVPDRFSGSGAGTDFTLKISRVEAEDVGVYYCVQATQFPLTFGGGTKVEIK
69	1.5.3 LCDR1	RSSQSLVYSDGNTYLS
70	1.5.3 LCDR2	KISNRFS
71	1.5.3 LCDR3	VQATQFPLT
72	human IgM constant region DNA	GCCCCAACCCTTTTCCCCCTCGTCTCCTGTGAGAATTCCCCGTCGGATACGAGCAGCG TGGCCGTTGGCTGCCTCGCACAGGACTTCCTTCCCGACTCCATCACTTTCTCCTGGAA ATACAAGAACAACTCTGACATCAGCAGCACCCGGGGCTTCCCATCAGTCCTGAGAGGG GGCAAGCACGCAGCCACCTCACAGGTGCTGCTGCCTTCCAAGGACGTCATGCAGGGCA CAGACGAACACGTGGTGTGCAAAGTCCAGCACCCCAACGGCAACAAAGAAAAGAACGT GCCTCTTCCAGTGATTGCTGAGCTGCCTCCCAAAGTGAGCGTCTTCGTCCCACCCCGC GACGGCTTCTTCGGCAACCCCCGCAAGTCCAAGCTCATCTGCCAGGCCACGGGTTTCA GTCCCCGGCAGATTCAGGTGTCCTGGCTGCGCGAGGGGAAGCAGGTGGGGTCTGGCGT CACCACGGACCAGGTGCAGGCTGAGGCAAAGGAGTCTGGGACCACGACCTACAAGGTG ACCAGCACACTGACCATCAAAGAGAGCGACTGGCTCAGCCAGAGCATGTTCACCTGCC GCGTGGATCACAGGGGCCTGACCTTCCAGCAGAATGCGTCCTCCATGTGTGGCCCCGA TCAAGACACAGCCATCCGGGTCTTCTCCATCCCCCCATCCTTTGCCAGCATCTTCCTC ACCAAGTCCACCAAGTTGACCTGCCTGGTCACAGACCTGACCACCTATGACAGCGTGA CCATCTCCTGGACCCGCCAGAATGGCGAAGCTGTGAAAACCCACACCAACATCTCCGA GAGCCACCCCAATGCCACTTTCAGCGCCGTGGGTGAGGCCAGCATCTGCGAGGATGAC TGGAATTCCGGGGAGAGGTTCACGTGCACCGTGACCCACACAGACCTGCCCTCGCCAC TGAAGCAGACCATCTCCCGGCCCAAGGGGGTGGCCCTGCACAGGCCCGATGTCTACTT GCTGCCACCAGCCCGGGAGCAGCTGAACCTGCGGGAGTCGGCCACCATCACGTGCCTG GTGACGGGCTTCTCTCCCGCGGACGTCTTCGTGCAGTGGATGCAGAGGGGGCAGCCCT TGTCCCCCGGAGAAGTATGTGACCAGCGCCCCAATGCCTGAGCCCCAGGCCCCACGCCG GTACTTCGCCCACAGCATCCTGACCGTGTCCGAAGAGGAATGGAACACGGGGGAGACC TACACCTGCGTGGTGGCCCATGAGGCCCTGCCCAACAGGGTCACCGAGAGGACCGTGG ACAAGTCCACCGGTAAACCCACCCTGTACAACGTGTCCCTGGTCATGTCCGACACAGC TGGCACCTGCTAC
73	human IgM constant region AA	GSASAPTLFPLVSCENSPSDTSSVAVGCLAQDFLPDSITFSWKYKNNSDISSTRGFPS VLRGGKYAATSQVLLPSKDVMQGTDEHVVCKVQHPNGNKEKNVPLPVIAELPPKVSVF VPPRDGFFGNPRKSKLICQATGFSPRQIQVSWLREGKQVGSGVTTDQVQAEAKESGPT TYKVTSTLTIKESDWLSQSMFTCRVDHRGLTFQQNASSMCVPDQDTAIRVFAIPPSFA SIFLTKSTKLTCLVTDLTTYDSVTISWTRQNGEAVKTHTNISESHPNATFSAVGEASI CEDDWNSGERFTCTVTHTDLPSPLKQTISRPKGVALHRPDVYLLPPAREQLNLRESAT ITCLVTGFSPADVFVQWMQRGQPLSPEKYVTSAPMPEPQAPGRYFAHSILTVSEEEWN TGETYTCVVAHEALPNRVTERTVDKSTGKPTLYNVSLVMSDTAGTCY

SEQ ID NO:	Short Name	Sequence
74	J Chain DNA	ATGAAGAACCATTTGCTTTTCTGGGGAGTCCTGGCGGTTTTTATTAAGGCTGTTCATG TGAAAGCCCAAGAAGATGAAAGGATTGTTCTTGTTGACAACAAATGTAAGTGTGCCCG GATTACTTCCAGGATCATCCGTTCTTCCGAAGATCCTAATGAGGACATTGTGGAGAGA AACATCCGAATTATTGTTCCTCTGAACAACAGGGAGAATATCTCTGATCCCACCTCAC CATTGAGAACCAGATTTGTGTACCATTTGTCTGACCTCTGTAAAAAATGTGATCCTAC AGAAGTGGAGCTGGATAATCAGATAGTTACTGCTACCCAGAGCAATATCTGTGATGAA GACAGTGCTACAGAGACCTGCTACACTTATGACAGAAACAAGTGCTACACAGCTGTGG TCCCACTCGTATATGGTGGTGAGACCAAAATGGTGGAAACAGCCTTAACCCCAGATGC CTGCTATCCTGACTAA
75	J Chain AA	MKNHLLFWGVLAVFIKAVHVKAQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVER NIIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDED SATETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPD
76	human CD20 amino acid	MTTPRNSVNGTFPAEPMKGPIAMQSGPKPLFRRMSSLVGPTQSFFMRESKTLGAVQIM NGLFHIALGGLLMIPAGIYAPICVTVWYPLWGGIMYIISGSLLAATEKNSRKCLVKGK MIMNSLSLFAAISGMILSIMDILNIKISHFLKMESLNFIRAHTPYINIYNCEPANPSE KNSPSTQYCYSIQSLFLGILSVMLIFAFFQELVIAGIVENEWKRTCSRPKSNIVLLSA EEKKEQTIEIKEEVVGLTETSSQPKNEEDIEIIPIQEEEEEETETNFPEPPQDQESSP IENDSSP
77	Ritux-IgM heavy chain DNA	CAGGTTCAGCTGCAGCAGCCCGGAGCCGAGCTGGTCAAACCTGGCGCTAGTGTGAAAA TGTCATGCAAGGCATCCGGATACACATTCACTAGCTATAACATGCACTGGGTGAAGCA GACCCCCGGCAGGGGTCTGGAGTGGATCGGAGCTATCTACCCCGGCAACGGAGACACA TCTTATAATCAGAAGTTTAAAGGCAAGGCCACCCTGACAGCTGATAAGTCCAGCTCTA CCGCATACATGCAGCTGAGTTCACTGACAAGCGAGGACTCCGCCGTGTACTATTGCGC CCGGTCCACTTACTATGGCGGAGATTGGTATTTCAATGTGTGGGGAGCAGGCACCACA GTCACCGTCTCGAGCGGCAGTGCTAGCGCCCCAACCCTTTTCCCCCTCGTCTCCTGTG AGAATTCCCCGTCGGATACGAGCAGCGTGGCCGTTGGCTGCCTCGCACAGGACTTCCT TCCCGACTCCATCACTTTCTCCTGGAAATACAAGAACAACTCTGACATCAGCAGCACC CGGGGCTTCCCATCAGTCCTGAGAGGGGGCAAGTACGCAGCCACCTCACAGGTGCTGC TGCCTTCCAAGGACGTCATGCAGGGCACAGACGAACACGTGGTGTGCAAAGTCCAGCA CCCCAACGGCAACAAAGAAAAGAACGTGCCTCTTCCAGTGATTGCTGAGCTGCCTCCC AAAGTGAGCGTCTTCGTCCCACCCCGCGACGGCTTCTTCGGCAACCCCCGCAAGTCCA AGCTCATCTGCCAGGCCACGGGTTTCAGTCCCCGGCAGATTCAGGTGTCCTGGCTGCG CGAGGGGAAGCAGGTGGGGTCTGGCGTCACCACGGACCAGGTGCAGGCTGAGGCCAAA GAGTCTGGGCCCACGACCTACAAGGTGACCAGCACACTGACCATCAAAGAGAGCGACT GGCTCAGCCAGAGCATGTTCACCTGCCGCGTGGATCACAGGGGCCTGACCTTCCAGCA GAATGCGTCCTCCATGTGTGTCCCCGATCAAGACACAGCCATCCGGGTCTTCGCCATC CCCCCATCCTTTGCCAGCATCTTCCTCACCAAGTCCACCAAGTTGACCTGCCTGGTCA CAGACCTGACCACCTATGACAGCGTGACCATCTCCTGGACCCGCCAGAATGGCGAAGC TGTGAAAACCCACACCAACATCTCCGAGAGCCACCCCAATGCCACTTTCAGCGCCGTG GGTGAGGCCAGCATCTGCGAGGATGACTGGAATTCCGGGGAGAGGTTCACGTGCACCG TGACCCACACAGACCTGCCCTCGCCACTGAAGCAGACCATCTCCCGGCCCAAGGGGGT GGCCCTGCACAGGCCCGATGTCTACTTGCTGCCACCAGCCCGGGAGCAGCTGAACCTG CGGGAGTCGGCCACCATCACGTGCCTGGTGACGGGCTTCTCTCCCGCGGACGTCTTCG TGCAGTGGATGCAGAGGGGGCAGCCCTTGTCCCCGGAGAAGTATGTGACCAGCGCCCC AATGCCTGAGCCCCAGGCCCCAGGCCGGTACTTCGCCCACAGCATCCTGACCGTGTCC GAAGAGGAATGGAACACGGGGGAGACCTACACCTGCGTGGTGGCCCATGAGGCCCTGC CCAACAGGGTCACCGAGAGGACCGTGGACAAGTCCACCGGTAAACCCACCCTGTACAA CGTGTCCCTGGTCATGTCCGACACAGCTGGCACCTGCTACTGA
78	Ritux-IgM heavy chain AA	QVQLQQPGAELVKPGASVKMSCKASGYTFTSYNMHWVKQTPGRGLEWIGAIYPGNGDT SYNQKFKGKATLTADKSSSTAYMQLSSLTSEDSAVYYCARSTYYGGDWYFNVWGAGTT VTVSSGSASAPTLFPLVSCENSPSDTSSVAVGCLAQDFLPDSITFSWKYKNNSDISST RGFPSVLRGGKYAATSQVLLPSKDVMQGTDEHVVCKVQHPNGNKEKNVPLPVIAELPP KVSVFVPPRDGFFGNPRKSKLICQATGFSPRQIQVSWLREGKQVGSGVTTDQVQAEAK ESGPTTYKVTSTLTIKESDWLSQSMFTCRVDHRGLTFQQNASSMCVPDQDTAIRVFAI

SEQ ID NO:	Short Name	Sequence
		PPSFASIFLTKSTKLTCLVTDLTTYDSVTISWTRQNGEAVKTHTNISESHPNATFSAV GEASICEDDWNSGERFTCTVTHTDLPSPLKQTISRPKGVALHRPDVYLLPPAREQLNL RESATITCLVTGFSPADVFVQWMQRGQPLSPEKYVTSAPMPEPQAPGRYFAHSILTVS EEEWNTGETYTCVVAHEALPNRVTERTVDKSTGKPTLYNVSLVMSDTAGTCY-
79	Ritux-light chain DNA	CAAATTGTGCTGTCTCAGAGTCCAGCTATCCTGAGCGCATCTCCCGGAGAGAAGGTGA CCATGACATGCAGAGCCTCCAGCTCTGTCTCCTACATCCACTGGTTCCAGCAGAAGCC CGGCTCCTCCCCAAAACCCTGGATCTACGCCACCTCTAACCTGGCTAGTGGTGTGCCT GTCAGGTTTAGTGGATCAGGGTCCGGCACCAGCTACTCTCTGACAATCAGCCGGGTGG AGGCTGAAGACGCCGCTACATACTATTGCCAGCAGTGGACTTCTAATCCCCCTACCTT CGGCGGAGGGACAAAGCTGGAGATCAAGCGTACGGTGGCTGCACCATCTGTCTTCATC TTCCCGCCATCTGATGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGA ATAACTTCTATCCCAGAGAGGCCAAAGTACAGTGGAAGGTGGATAACGCCCTCCAATC GGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCACCTACAGCCTC AGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCG AAGTCACCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTG TTAG
80	Ritux-light chain AA	QIVLSQSPAILSASPGEKVTMTCRASSSVSYIHWFQQKPGSSPKPWIYATSNLASGVP VRFSGSGSGTSYSLTISRVEAEDAATYYCQQWTSNPPTFGGGTKLEIKRTVAAPSVFI FPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSL SSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC-
81	1.5.3 -IgM heavy chain DNA	GAGGTGCAGCTGGTGCAGTCCGGCGCCGAGGTGAAGAAGCCCGGCGAGTCCCTGAAGA TCTCCTGCAAGGGCTCCGGCTACTCCTTCACCTCCTACTGGATCGGCTGGGTGAGGCA GATGCCCGGCAAGGGCCTGGAGTGGATGGGCATCATCTACCCCGGCGACTCCGACACC AGGTACTCCCCCTCCTTCCAGGGCCAGGTGACCATCTCCGCCGACAAGTCCATCACCA CCGCCTACCTGCAGTGGTCCTCCCTGAAGGCCTCCGACACCGCCATGTACTACTGCGC CAGGCACCCCTCCTACGGCTCCGGCTCCCCCAACTTCGACTACTGGGGCCAGGGCACC CTGGTGACCGTGTCCTCCGGCAGTGCTAGCGCCCCAACCCTTTTCCCCCTCGTCTCCT GTGAGAATTCCCCGTCGGATACGAGCAGCGTGGCCGTTGGCTGCCTCGCACAGGACTT CCTTCCCGACTCCATCACTTTCTCCTGGAAATACAAGAACAACTCTGACATCAGCAGC ACCCGGGGCTTCCCATCAGTCCTGAGAGGGGGCAAGTACGCAGCCACCTCACAGGTGC TGCTGCCTTCCAAGGACGTCATGCAGGGCACAGACGAACACGTGGTGTGCAAAGTCCA GCACCCCAACGGCAACAAAGAAAAGAACGTGCCTCTTCCAGTGATTGCTGAGCTGCCT CCCAAAGTGAGCGTCTTCGTCCCACCCCGCGACGGCTTCTTCGGCAACCCCCGCAAGT CCAAGCTCATCTGCCAGGCCACGGGTTTCAGTCCCCGGCAGATTCAGGTGTCCTGGCT GCGCGAGGGGAAGCAGGTGGGGTCTGGCGTCACCACGGACCAGGTGCAGGCTGAGGCC AAAGAGTCTGGGCCCACGACCTACAAGGTGACCAGCACACTGACCATCAAAGAGAGCG ACTGGCTCAGCCAGAGCATGTTCACCTGCCGCGTGGATCACAGGGGCCTGACCTTCCA GCAGAATGCGTCCTCCATGTGTGTCCCCGATCAAGACACAGCCATCCGGGTCTTCGCC ATCCCCCCATCCTTTGCCAGCATCTTCCTCACCAAGTCCACCAAGTTGACCTGCCTGG TCACAGACCTGACCACCTATGACAGCGTGACCATCTCCTGGACCCGCCAGAATGGCGA AGCTGTGAAAACCCACACCAACATCTCCGAGAGCCACCCCAATGCCACTTTCAGCGCC GTGGGTGAGGCCAGCATCTGCGAGGATGACTGGAATTCCGGGGAGAGGTTCACGTGCA CCGTGACCCACACAGACCTGCCCTCGCCACTGAAGCAGACCATCTCCCGGCCCAAGGG GGTGGCCCTGCACAGGCCCGATGTCTACTTGCTGCCACCAGCCCGGGAGCAGCTGAAC CTGCGGGAGTCGGCCACCATCACGTGCCTGGTGACGGGCTTCTCTCCCGCGGACGTCT TCGTGCAGTGGATGCAGAGGGGGCAGCCCTTGTCCCCGGAGAAGTATGTGACCAGCGC CCCAATGCCTGAGCCCCAGGCCCCAGGCCGGTACTTCGCCCACAGCATCCTGACCGTG TCCGAAGAGGAATGGAACACGGGGGAGACCTACACCTGCGTGGTGGCCCATGAGGCCC TGCCCAACAGGGTCACCGAGAGGACCGTGGACAAGTCCACCGGTAAACCCACCCTGTA CAACGTGTCCCTGGTCATGTCCGACACAGCTGGCACCTGCTACTGA

SEQ ID NO:	Short Name	Sequence
82	1.5.3 -IgM heavy chain AA	EVQLVQSGAEVKKPGESLKISCKGSGYSFTSYWIGWVRQMPGKGLEWMGIIYPGDSDT RYSPSFQGQVTISADKSITTAYLQWSSLKASDTAMYYCARHPSYGSGSPNFDYWGQGT LVTVSSGSASAPTLFPLVSCENSPSDTSSVAVGCLAQDFLPDSITFSWKYKNNSDISS TRGFPSVLRGGKYAATSQVLLPSKDVMQGTDEHVVCKVQHPNGNKEKNVPLPVIAELP PKVSVFVPPRDGFFGNPRKSKLICQATGFSPRQIQVSWLREGKQVGSGVTTDQVQAEA KESGPTTYKVTSTLTIKESDWLSQSMFTCRVDHRGLTFQQNASSMCVPDQDTAIRVFA IPPSFASIFLTKSTKLTCLVTDLTTYDSVTISWTRQNGEAVKTHTNISESHPNATFSA VGEASICEDDWNSGERFTCTVTHTDLPSPLKQTISRPKGVALHRPDVYLLPPAREQLN LRESATITCLVTGFSPADVFVQWMQRGQPLSPEKYVTSAPMPEPQAPGRYFAHSILTV SEEEWNTGETYTCVVAHEALPNRVTERTVDKSTGKPTLYNVSLVMSDTAGTCY-
83	1.5.3 light chain DNA	GACATCGTGATGACCCAGACCCCCCTGTCCTCCCCCGTGACCCTGGGCCAGCCCGCCT CCATCTCCTGCAGGTCCTCCCAGTCCCTGGTGTACTCCGACGGCAACACCTACCTGTC CTGGCTGCAGCAGAGGCCCGGCCAGCCCCCCAGGCTGCTGATCTACAAGATCTCCAAC AGGTTCTCCGGCGTGCCCGACAGGTTCTCCGGCTCCGGCGCCGGCACCGACTTCACCC TGAAGATCTCCAGGGTGGAGGCCGAGGACGTGGGCGTGTACTACTGCGTGCAGGCCAC CCAGTTCCCCCTGACCTTCGGCGGCGGCACCAAGGTGGAGATCAAGCGTACGGTGGCT GCACCATCTGTCTTCATCTTCCCGCCATCTGATGAGCAGTTGAAATCTGGAACTGCCT CTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGTGGAAGGT GGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAG GACAGCACCTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAAC ACAAAGTCTACGCCTGCGAAGTCACCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAG CTTCAACAGGGGAGAGTGTTAG
84	1.5.3 light chain AA	DIVMTQTPLSSPVTLGQPASISCRSSQSLVYSDGNTYLSWLQQRPGQPPRLLIYKISN RFSGVPDRFSGSGAGTDFTLKISRVEAEDVGVYYCVQATQFPLTFGGGTKVEIKRTVA APSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSK DSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC-
85	human IgA1 constant region aa P01876	ASPTSPKVFPLSLCSTQPDGNVVIACLVQGFFPQEPLSVTWSESGQGVTARNFPPSQD ASGDLYTTSSQLTLPATQCLAGKSVTCHVKHYTNPSQDVTVPCPVPSTPPTPSPSTPP TPSPSCCHPRLSLHRPALEDLLLGSEANLTCTLTGLRDASGVTFTWTPSSGKSAVQGP PERDLCGCYSVSSVLPGCAEPWNHGKTFTCTAAYPESKTPLTATLSKSGNTFRPEVHL LPPPSEELALNELVTLTCLARGFSPKDVLVRWLQGSQELPREKYLTWASRQEPSQGTT TFAVTSILRVAAEDWKKGDTFSCMVGHEALPLAFTQKTIDRLAGKPTHVNVSVVMAEV DGTCY
86	human IgA2 constant region aa P01877	ASPTSPKVFPLSLDSTPQDGNVVVACLVQGFFPQEPLSVTWSESGQNVTARNFPPSQD ASGDLYTTSSQLTLPATQCPDGKSVTCHVKHYTNPSQDVTVPCPVPPPPPCCHPRLSL HRPALEDLLLGSEANLTCTLTGLRDASGATFTWTPSSGKSAVQGPPERDLCGCYSVSS VLPGCAQPWNHGETFTCTAAHPELKTPLTANITKSGNTFRPEVHLLPPPSEELALNEL VTLTCLARGFSPKDVLVRWLQGSQELPREKYLTWASRQEPSQGTTTFAVTSILRVAAE DWKKGDTFSCMVGHEALPLAFTQKTIDRMAGKPTHVNVSVVMAEVDGTCY
87	Human Secretory Component Precursor	MLLFVLTCLLAVFPAISTKSPIFGPEEVNSVEGNSVSITCYYPPTSVNRHTRKYWCRQ GARGGCITLISSEGYVSSKYAGRANLTNFPENGTFVVNIAQLSQDDSGRYKCGLGINS RGLSFDVSLEVSQGPGLLNDTKVYTVDLGRTVTINCPFKTENAQKRKSLYKQIGLYPV LVIDSSGYVNPNYTGRIRLDIQGTGQLLFSVVINQLRLSDAGQYLCQAGDDSNSNKKN ADLQVLKPEPELVYEDLRGSVTFHCALGPEVANVAKFLCRQSSGENCDVVVNTLGKRA PAFEGRILLNPQDKDGSFSVVITGLRKEDAGRYLCGAHSDGQLQEGSPIQAWQLFVNE ESTIPRSPTVVKGVAGGSVAVLCPYNRKESKSIKYWCLWEGAQNGRCPLLVDSEGWVK AQYEGRLSLLEEPGNGTFTVILNQLTSRDAGFYWCLTNGDTLWRTTVEIKIIEGEPNL KVPGNVTAVLGETLKVPCHFPCKFSSYEKYWCKWNNTGCQALPSQDEGPSKAFVNCDE NSRLVSLTLNLVTRADEGWYWCGVKQGHFYGETAAVYVAVEERKAAGSRDVSLAKADA APDEKVLDSGFREIENKAIQDPRLFAEEKAVADTRDQADGSRASVDSGSSEEQGGSSR ALVSTLVPLGLVLAVGAVAVGVARARHRKNVDRVSIRSYRTDISMSDFENSREFGAND NMGASSITQETSLGGKEEFVATTESTTETKEPKKAKRSSKEEAEMAYKDFLLQSSTVA AEAQDGPQEA

SEQ ID NO:	Short Name	Sequence
88	human secretory component mature	KSPIFGPEEVNSVEGNSVSITCYYPPTSVNRHTRKYWCRQGARGGCITLISSEGYVSS KYAGRANLTNFPENGTFVVNIAQLSQDDSGRYKCGLGINSRGLSFDVSLEVSQGPGLL NDTKVYTVDLGRTVTINCPFKTENAQKRKSLYKQIGLYPVLVIDSSGYVNPNYTGRIR LDIQGTGQLLFSVVINQLRLSDAGQYLCQAGDDSNSNKKNADLQVLKPEPELVYEDLR GSVTFHCALGPEVANVAKFLCRQSSGENCDVVVNTLGKRAPAFEGRILLNPQDKDGSF SVVITGLRKEDAGRYLCGAHSDGQLQEGSPIQAWQLFVNEESTIPRSPTVVKGVAGGS VAVLCPYNRKESKSIKYWCLWEGAQNGRCPLLVDSEGWVKAQYEGRLSLLEEPGNGTF TVILNQLTSRDAGFYWCLTNGDTLWRTTVEIKIIEGEPNLKVPGNVTAVLGETLKVPC HFPCKFSSYEKYWCKWNNTGCQALPSQDEGPSKAFVNCDENSRLVSLTLNLVTRADEG WYWCGVKQGHFYGETAAVYVAVEERKAAGSRDVSLAKADAAPDEKVLDSGFREIENKA IQDPR
89	J15ABD DNA	ATGGAATGGAGCTGGGTCTTTCTCTTCTTCCTGTCAGTAACGACTGGTGTCCACTCCC AGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAGTGCGCCCGGATCACCTC CCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCGTGGAACGGAACATCAGA ATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCCCACCAGCCCTCTGCGGA CCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGCGACCCTACCGAGGTGGA ACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCTGCGACGAGGACTCCGCC ACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACACCGCCGTGGTGCCTCTGG TGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACCCCCGACGCCTGCTATCC TGATGGAGGCGGAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGGGCTCTCAGCACGAT GAGGCCGTGGACGCCAATTCTCTGGCCGAGGCTAAGGTGCTGGCCAACAGAGAGCTGG ATAAGTACGGCGTGTCCGACTACTACAAGAACCTGATCAACAACGCCAAGACCGTGGA AGGCGTGAAGGCCCTGATCGACGAGATCCTGGCTGCCCTGCCTTGA
90	J15ABD AA	MEWSWVFLFFLSVTTGVHSQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNIR IIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDEDSA TETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPDGGGGSGGGGSGGGGSQHD EAVDANSLAEAKVLANRELDKYGVSDYYKNLINNAKTVEGVKALIDEILAALP
91	ABD15J DNA	ATGGAATGGAGCTGGGTCTTTCTCTTCTTCCTGTCAGTAACGACTGGTGTCCACTCCC AGCACGATGAGGCCGTGGACGCCAATTCTCTGGCCGAGGCTAAGGTGCTGGCCAACAG AGAGCTGGATAAGTACGGCGTGTCCGACTACTACAAGAACCTGATCAACAACGCCAAG ACCGTGGAAGGCGTGAAGGCCCTGATCGACGAGATCCTGGCTGCCCTGCCTGGAGGCG GAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGGGCTCTCAGGAAGATGAGCGGATCGT GCTGGTGGACAACAAGTGCAAGTGCGCCCGGATCACCTCCCGGATCATCCGGTCCTCC GAGGATCCCAACGAGGACATCGTGGAACGGAACATCAGAATCATCGTGCCCCTGAACA ACCGCGAGAACATCTCCGACCCCACCAGCCCTCTGCGGACCAGATTCGTGTACCACCT GTCCGACCTGTGCAAGAAGTGCGACCCTACCGAGGTGGAACTGGACAACCAGATCGTG ACCGCCACCCAGTCCAACATCTGCGACGAGGACTCCGCCACCGAGACATGCTACACCT ACGACCGGAACAAGTGCTACACCGCCGTGGTGCCTCTGGTGTACGGCGGCGAGACAAA GATGGTGGAAACCGCCCTGACCCCCGACGCCTGCTATCCTGATTGA
92	ABD15J AA	MEWSWVFLFFLSVTTGVHSQHDEAVDANSLAEAKVLANRELDKYGVSDYYKNLINNAK TVEGVKALIDEILAALPGGGGSGGGGSGGGGSQEDERIVLVDNKCKCARITSRIIRSS EDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIV TATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPD
93	HSA15J DNA	ATGAAATGGGTCACCTTTATCTCCCTGCTGTTCCTGTTCTCCTCCGCCTACTCTCGGG GCGTGTTCAGAAGAGACGCCCACAAATCGGAGGTAGCGCACCGGTTCAAAGACTTGGG AGAAGAAAACTTTAAGGCCCTTGTACTCATTGCGTTTGCGCAGTATTTGCAGCAGTGC CCATTCGAGGACCATGTCAAACTTGTCAACGAAGTGACAGAGTTTGCGAAAACTTGCG TCGCCGACGAATCCGCGGACAACTGTGACAAGTCGCTGCATACGTTGTTCGGGGATAA GCTCTGTACCGTAGCGACCTTGAGGGAAACTTACGGGGAAATGGCGGACTGTTGCGCT AAGCAGGAGCCGGAACGGAACGAGTGTTTCCTTCAGCATAAGGATGACAACCCCAACC TCCCTAGATTGGTCAGACCCGAAGTGGATGTGATGTGCACAGCATTCCATGACAATGA GGAAACCTTTCTCAAAAAGTATTTGTACGAGATTGCCCGACGACACCCCTATTTCTAC GCTCCCGAGTTGCTCTTCTTCGCGAAACGGTATAAAGCTGCCTTTACTGAATGCTGTC AAGCAGCGGACAAGGCCGCATGCCTCCTTCCCAAATTGGATGAACTCCGCGATGAAGG

SEQ ID NO:	Short Name	Sequence
		GAAGGCGTCATCGGCCAAACAGCGGCTTAAGTGCGCATCGCTTCAGAAATTCGGAGAG AGGGCGTTCAAAGCGTGGGCCGTCGCGAGACTGTCGCAGAGATTCCCTAAGGCGGAAT TTGCAGAGGTATCGAAGCTCGTGACAGACCTCACAAAGGTCCACACCGAATGTTGCCA TGGAGACCTGCTTGAGTGCGCCGATGATAGGGCAGACCTCGCAAAGTACATTTGTGAG AATCAGGACAGCATTAGCTCCAAGCTGAAAGAGTGCTGTGAGAAGCCTTTGCTGGAAA AATCCCACTGTATCGCCGAGGTAGAAAACGATGAAATGCCCGCTGATCTTCCCTCGCT GGCGGCAGACTTCGTCGAGTCGAAGGACGTCTGCAAGAATTACGCAGAGGCAAAAGAT GTGTTTCTTGGAATGTTCCTTTATGAGTATGCGAGAAGGCACCCGGATTATTCCGTGG TACTGCTCTTGCGATTGGCGAAAACGTACGAAACAACGCTTGAGAAGTGTTGTGCGGC TGCCGACCCGCATGAGTGCTACGCCAAGGTATTTGATGAGTTTAAACCTCTTGTCGAG GAACCCCAGAATCTTATCAAGCAGAACTGCGAGCTTTTCAAGCAGTTGGGTGAATACA AATTCCAGAACGCGCTTCTGGTGAGGTATACCAAGAAAGTACCTCAAGTCTCAACACC CACACTCGTCGAGGTGTCACGGAACCTCGGGAAAGTAGGGTCGAAGTGCTGTAAACAC CCAGAGGCCAAGCGCATGCCCTGTGCGGAGGACTACCTCTCGGTAGTGTTGAATCAAC TGTGTGTCCTCCACGAAAAGACGCCGGTGTCAGACCGCGTCACAAAGTGCTGCACGGA GAGCCTGGTCAATAGACGCCCCTGCTTCTCAGCGCTGGAGGTGGATGAGACATACGTC CCGAAAGAGTTTAACGCCGAAACGTTTACTTTTCATGCTGATATCTGTACGTTGTCAG AGAAGGAAAGGCAAATCAAGAAACAAACTGCGCTTGTGGAACTGGTGAAGCACAAACC GAAGGCGACTAAGGAACAGCTGAAGGCGGTGATGGATGACTTTGCCGCGTTCGTAGAG AAATGCTGTAAAGCAGACGATAAGGAGACTTGTTTTGCGGAAGAGGGACCTAAACTTG TTGCTGCAAGTCAAGCTGCCTTAGGCTTAGGAGGCGGAGGATCTGGTGGCGGTGGTTC TGGCGGAGGGGGCTCTCAGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAG TGCGCCCGGATCACCTCCCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCG TGGAACGGAACATCAGAATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCC CACCAGCCCTCTGCGGACCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGC GACCCTACCGAGGTGGAACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCT GCGACGAGGACTCCGCCACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACAC CGCCGTGGTGCCTCTGGTGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACC CCCGACGCCTGCTATCCTGATTAG
94	HSA15J AA	MKWVTFISLLFLFSSAYSRGVFRRDAHKSEVAHRFKDLGEENFKALVLIAFAQYLQQC PFEDHVKLVNEVTEFAKTCVADESAENCDKSLHTLFGDKLCTVATLRETYGEMADCCA KQEPERNECFLQHKDDNPNLPRLVRPEVDVMCTAFHDNEETFLKKYLYEIARRHPYFY APELLFFAKRYKAAFTECCQAADKAACLLPKLDELRDEGKASSAKQRLKCASLQKFGE RAFKAWAVARLSQRFPKAEFAEVSKLVTDLTKVHTECCHGDLLECADDRADLAKYICE NQDSISSKLKECCEKPLLEKSHCIAEVENDEMPADLPSLAADFVESKDVCKNYAEAKD VFLGMFLYEYARRHPDYSVVLLLRLAKTYETTLEKCCAAADPHECYAKVFDEFKPLVE EPQNLIKQNCELFKQLGEYKFQNALLVRYTKKVPQVSTPTLVEVSRNLGKVGSKCCKH PEAKRMPCAEDYLSVVLNQLCVLHEKTPVSDRVTKCCTESLVNRRPCFSALEVDETYV PKEFNAETFTFHADICTLSEKERQIKKQTALVELVKHKPKATKEQLKAVMDDFAAFVE KCCKADDKETCFAEEGPKLVAASQAALGLGGGGSGGGGSGGGGSQEDERIVLVDNKCK CARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKC DPTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALT PDACYPD
95	J15HSA DNA	ATGAAGAACCATCTGCTGTTCTGGGGCGTGCTGGCCGTGTTCATCAAGGCCGTGCACG TGAAGGCCCAGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAGTGCGCCCG GATCACCTCCCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCGTGGAACGG AACATCAGAATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCCCACCAGCC CTCTGCGGACCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGCGACCCTAC CGAGGTGGAACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCTGCGACGAG GACTCCGCCACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACACCGCCGTGG TGCCTCTGGTGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACCCCCGACGC CTGCTATCCTGATGGAGGCGGAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGGGCTCT GACGCCCACAAATCGGAGGTAGCGCACCGGTTCAAAGACTTGGGAGAAGAAAACTTTA AGGCCCTTGTACTCATTGCGTTTGCGCAGTATTTGCAGCAGTGCCCATTCGAGGACCA TGTCAAACTTGTCAACGAAGTGACAGAGTTTGCGAAAACTTGCGTCGCCGACGAATCC

SEQ ID NO:	Short Name	Sequence
		GCGGAGAACTGTGACAAGTCGCTGCATACGTTGTTCGGGGATAAGCTCTGTACCGTAG CGACCTTGAGGGAAACTTACGGGGAAATGGCGGACTGTTGCGCTAAGCAGGAGCCGGA ACGGAACGAGTGTTTCCTTCAGCATAAGGATGACAACCCCAACCTCCCTAGATTGGTC AGACCCGAAGTGGATGTGATGTGCACAGCATTCCATGACAATGAGGAAACCTTTCTCA AAAAGTATTTGTACGAGATTGCCCGACGACACCCCTATTTCTACGCTCCCGAGTTGCT CTTCTTCGCGAAACGGTATAAAGCTGCCTTTACTGAATGCTGTCAAGCAGCGGACAAG GCCGCATGCCTCCTTCCCAAATTGGATGAACTCCGCGATGAAGGGAAGGCGTCATCGG CCAAACAGCGGCTTAAGTGCGCATCGCTTCAGAAATTCGGAGAGAGGGCGTTCAAAGC GTGGGCCGTCGCGAGACTGTCGCAGAGATTCCCTAAGGCGGAATTTGCAGAGGTATCG AAGCTCGTGACAGACCTCACAAAGGTCCACACCGAATGTTGCCATGGAGACCTGCTTG AGTGCGCCGATGATAGGGCAGACCTCGCAAAGTACATTTGTGAGAATCAGGACAGCAT TAGCTCCAAGCTGAAAGAGTGCTGTGAGAAGCCTTTGCTGGAAAAATCCCACTGTATC GCCGAGGTAGAAAACGATGAAATGCCCGCTGATCTTCCCTCGCTGGCGGCAGACTTCG TCGAGTCGAAGGACGTCTGCAAGAATTACGCAGAGGCAAAAGATGTGTTTCTTGGAAT GTTCCTTTATGAGTATGCGAGAAGGCACCCGGATTATTCCGTGGTACTGCTCTTGCGA TTGGCGAAAACGTACGAAACAACGCTTGAGAAGTGTTGTGCGGCTGCCGACCCGCATG AGTGCTACGCCAAGGTATTTGATGAGTTTAAACCTCTTGTCGAGGAACCCCAGAATCT TATCAAGCAGAACTGCGAGCTTTTCAAGCAGTTGGGTGAATACAAATTCCAGAACGCG CTTCTGGTGAGGTATACCAAGAAAGTACCTCAAGTCTCAACACCCACACTCGTCGAGG TGTCACGGAACCTCGGGAAAGTAGGGTCGAAGTGCTGTAAACACCCAGAGGCCAAGCG CATGCCCTGTGCGGAGGACTACCTCTCGGTAGTGTTGAATCAACTGTGTGTCCTCCAC GAAAAGACGCCGGTGTCAGACCGCGTCACAAAGTGCTGCACGGAGAGCCTGGTCAATA GACGCCCCTGCTTCTCAGCGCTGGAGGTGGATGAGACATACGTCCCGAAAGAGTTTAA CGCCGAAACGTTTACTTTTCATGCTGATATCTGTACGTTGTCAGAGAAGGAAAGGCAA ATCAAGAAACAAACTGCGCTTGTGGAACTGGTGAAGCACAAACCGAAGGCGACTAAGG AACAGCTGAAGGCGGTGATGGATGACTTTGCCGCGTTCGTAGAGAAATGCTGTAAAGC AGACGATAAGGAGACTTGTTTTGCGGAAGAGGGACCTAAACTTGTTGCTGCAAGTCAA GCTGCCTTAGGCTTATAG
96	J15HSAAA	MKNHLLFWGVLAVFIKAVHVKAQEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVER NIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPTEVELDNQIVTATQSNICDE DSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTPDACYPDGGGGSGGGGSGGGGS DAHKSEVAHRFKDLGEENFKALVLIAFAQYLQQCPFEDHVKLVNEVTEFAKTCVADES AENCDKSLHTLFGDKLCTVATLRETYGEMADCCAKQEPERNECFLQHKDDNPNLPRLV RPEVDVMCTAFHDNEETFLKKYLYEIARRHPYFYAPELLFFAKRYKAAFTECCQAADK AACLLPKLDELRDEGKASSAKQRLKCASLQKFGERAFKAWAVARLSQRFPKAEFAEVS KLVTDLTKVHTECCHGDLLECADDRADLAKYICENQDSISSKLKECCEKPLLEKSHCI AEVENDEMPADLPSLAADFVESKDVCKNYAEAKDVFLGMFLYEYARRHPDYSVVLLLR LAKTYETTLEKCCAAADPHECYAKVFDEFKPLVEEPQNLIKQNCELFKQLGEYKFQNA LLVRYTKKVPQVSTPTLVEVSRNLGKVGSKCCKHPEAKRMPCAEDYLSVVLNQLCVLH EKTPVSDRVTKCCTESLVNRRPCFSALEVDETYVPKEFNAETFTFHADICTLSEKERQ IKKQTALVELVKHKPKATKEQLKAVMDDFAAFVEKCCKADDKETCFAEEGPKLVAASQ AALGL
97	V15J15ABDDNA	ATGGGGTGGTCCTACATTATCCTGTTCCTCGTGGCCACCGCCACTGGCGTGCACTCAC AGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGGT GTCCTGCAAGGCCTCCGGCTACACCTTCATCAGCTACACCATGCACTGGGTGCGACAG GCCCCTGGACAGGGCCTGGAATGGATGGGCTACATCAACCCTAGATCTGGCTACACCC ACTACAACCAGAAGCTGAAGGACAAGGCCACCCTGACCGCCGACAAGTCTGCCTCCAC CGCCTACATGGAACTGTCCTCCCTGCGGAGCGAGGACACCGCCGTGTACTACTGTGCC AGATCCGCCTACTACGACTACGACGGCTTCGCCTATTGGGGCCAGGGCACCCTCGTGA CAGTGTCTAGCGGTGGCGGAGGATCTGGCGGAGGCGGTAGTGGCGGTGGCGGATCTGA TATCCAGATGACCCAGTCCCCCTCCAGCCTGTCTGCCTCTGTGGGCGACAGAGTGACA ATTACCTGCTCCGCCAGCTCCTCCGTGTCTTACATGAACTGGTATCAGCAGAAGCCCG GCAAGGCCCCCAAGCGGCTGATCTACGACACCTCCAAGCTGGCCTCTGGCGTGCCCTC CAGATTCTCCGGCTCTGGCTCTGGCACCGACTTTACCCTGACCATCAGCTCCCTGCAG CCCGAGGACTTCGCCACCTACTACTGCCAGCAGTGGTCCTCCAACCCTCCCACCTTTG

SEQ ID NO:	Short Name	Sequence
		GCGGAGGCACCAAGGTGGAAATCAAAGGCGGCGGAGGAAGCGGGGGAGGCGGTTCTGG GGGTGGTGGATCTCAGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAGTGC GCCCGGATCACCTCCCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCGTGG AACGGAACATCAGAATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCCCAC CAGCCCTCTGCGGACCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGCGAC CCTACCGAGGTGGAACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCTGCG ACGAGGACTCCGCCACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACACCGC CGTGGTGCCTCTGGTGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACCCCC GACGCCTGCTATCCTGATGGAGGCGGAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGG GCTCTCAGCACGATGAGGCCGTGGACGCCAATTCTCTGGCCGAGGCTAAGGTGCTGGC CAACAGAGAGCTGGATAAGTACGGCGTGTCCGACTACTACAAGAACCTGATCAACAAC GCCAAGACCGTGGAAGGCGTGAAGGCCCTGATCGACGAGATCCTGGCTGCCCTGCCTT GA
98	V15J15ABD AA	MGWSYIILFLVATATGVHSQVQLVQSGAEVKKPGASVKVSCKASGYTFISYTMHWVRQ APGQGLEWMGYINPRSGYTHYNQKLKDKATLTADKSASTAYMELSSLRSEDTAVYYCA RSAYYDYDGFAYWGQGTLVTVSSGGGGSGGGGSGGGGSDIQMTQSPSSLSASVGDRVT ITCSASSSVSYMNWYQQKPGKAPKRLIYDTSKLASGVPSRFSGSGSGTDFTLTISSLQ PEDFATYYCQQWSSNPPTFGGGTKVEIKGGGGSGGGGSGGGGSQEDERIVLVDNKCKC ARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCD PTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTP DACYPDGGGGSGGGGSGGGGSQHDEAVDANSLAEAKVLANRELDKYGVSDYYKNLINN AKTVEGVKALIDEILAALP
99	V15J15HSA(K573P) DNA	ATGGGGTGGTCCTACATTATCCTGTTCCTCGTGGCCACCGCCACTGGCGTGCACTCAC AGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGGT GTCCTGCAAGGCCTCCGGCTACACCTTCATCAGCTACACCATGCACTGGGTGCGACAG GCCCCTGGACAGGGCCTGGAATGGATGGGCTACATCAACCCTAGATCTGGCTACACCC ACTACAACCAGAAGCTGAAGGACAAGGCCACCCTGACCGCCGACAAGTCTGCCTCCAC CGCCTACATGGAACTGTCCTCCCTGCGGAGCGAGGACACCGCCGTGTACTACTGTGCC AGATCCGCCTACTACGACTACGACGGCTTCGCCTATTGGGGCCAGGGCACCCTCGTGA CAGTGTCTAGCGGTGGCGGAGGATCTGGCGGAGGCGGTAGTGGCGGTGGCGGATCTGA TATCCAGATGACCCAGTCCCCCTCCAGCCTGTCTGCCTCTGTGGGCGACAGAGTGACA ATTACCTGCTCCGCCAGCTCCTCCGTGTCTTACATGAACTGGTATCAGCAGAAGCCCG GCAAGGCCCCCAAGCGGCTGATCTACGACACCTCCAAGCTGGCCTCTGGCGTGCCCTC CAGATTCTCCGGCTCTGGCTCTGGCACCGACTTTACCCTGACCATCAGCTCCCTGCAG CCCGAGGACTTCGCCACCTACTACTGCCAGCAGTGGTCCTCCAACCCTCCCACCTTTG GCGGAGGCACCAAGGTGGAAATCAAAGGCGGCGGAGGAAGCGGGGGAGGCGGTTCTGG GGGTGGTGGATCTCAGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAGTGC GCCCGGATCACCTCCCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCGTGG AACGGAACATCAGAATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCCCAC CAGCCCTCTGCGGACCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGCGAC CCTACCGAGGTGGAACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCTGCG ACGAGGACTCCGCCACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACACCGC CGTGGTGCCTCTGGTGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACCCCC GACGCCTGCTATCCTGATGGAGGCGGAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGG GCTCTGACGCCCACAAATCGGAGGTAGCGCACCGGTTCAAAGACTTGGGAGAAGAAAA CTTTAAGGCCCTTGTACTCATTGCGTTTGCGCAGTATTTGCAGCAGTGCCCATTCGAG GACCATGTCAAACTTGTCAACGAAGTGACAGAGTTTGCGAAAACTTGCGTCGCCGACG AATCCGCGGAGAACTGTGACAAGTCGCTGCATACGTTGTTCGGGGATAAGCTCTGTAC CGTAGCGACCTTGAGGGAAACTTACGGGGAAATGGCGGACTGTTGCGCTAAGCAGGAG CCGGAACGGAACGAGTGTTTCCTTCAGCATAAGGATGACAACCCCAACCTCCCTAGAT TGGTCAGACCCGAAGTGGATGTGATGTGCACAGCATTCCATGACAATGAGGAAACCTT TCTCAAAAAGTATTTGTACGAGATTGCCCGACGACACCCCTATTTCTACGCTCCCGAG TTGCTCTTCTTCGCGAAACGGTATAAAGCTGCCTTTACTGAATGCTGTCAAGCAGCGG ACAAGGCCGCATGCCTCCTTCCCAAATTGGATGAACTCCGCGATGAAGGGAAGGCGTC ATCGGCCAAACAGCGGCTTAAGTGCGCATCGCTTCAGAAATTCGGAGAGAGGGCGTTC

SEQ ID NO:	Short Name	Sequence
		AAAGCGTGGGCCGTCGCGAGACTGTCGCAGAGATTCCCTAAGGCGGAATTTGCAGAGG TATCGAAGCTCGTGACAGACCTCACAAAGGTCCACACCGAATGTTGCCATGGAGACCT GCTTGAGTGCGCCGATGATAGGGCAGACCTCGCAAAGTACATTTGTGAGAATCAGGAC AGCATTAGCTCCAAGCTGAAAGAGTGCTGTGAGAAGCCTTTGCTGGAAAAATCCCACT GTATCGCCGAGGTAGAAAACGATGAAATGCCCGCTGATCTTCCCTCGCTGGCGGCAGA CTTCGTCGAGTCGAAGGACGTCTGCAAGAATTACGCAGAGGCAAAAGATGTGTTTCTT GGAATGTTCCTTTATGAGTATGCGAGAAGGCACCCGGATTATTCCGTGGTACTGCTCT TGCGATTGGCGAAAACGTACGAAACAACGCTTGAGAAGTGTTGTGCGGCTGCCGACCC GCATGAGTGCTACGCCAAGGTATTTGATGAGTTTAAACCTCTTGTCGAGGAACCCCAG AATCTTATCAAGCAGAACTGCGAGCTTTTCAAGCAGTTGGGTGAATACAAATTCCAGA ACGCGCTTCTGGTGAGGTATACCAAGAAAGTACCTCAAGTCTCAACACCCACACTCGT CGAGGTGTCACGGAACCTCGGGAAAGTAGGGTCGAAGTGCTGTAAACACCCAGAGGCC AAGCGCATGCCCTGTGCGGAGGACTACCTCTCGGTAGTGTTGAATCAACTGTGTGTCC TCCACGAAAAGACGCCGGTGTCAGACCGCGTCACAAAGTGCTGCACGGAGAGCCTGGT CAATAGACGCCCCTGCTTCTCAGCGCTGGAGGTGGATGAGACATACGTCCCGAAAGAG TTTAACGCCGAAACGTTTACTTTTCATGCTGATATCTGTACGTTGTCAGAGAAGGAAA GGCAAATCAAGAAACAAACTGCGCTTGTGGAACTGGTGAAGCACAAACCGAAGGCGAC TAAGGAACAGCTGAAGGCGGTGATGGATGACTTTGCCGCGTTCGTAGAGAAATGCTGT AAAGCAGACGATAAGGAGACTTGTTTTGCGGAAGAGGGACCTAAACTTGTTGCTGCAA GTCAAGCTGCCTTAGGCTTATAG
100	V15J15HSA(K573P) AA	MGWSYIILFLVATATGVHSQVQLVQSGAEVKKPGASVKVSCKASGYTFISYTMHWVRQ APGQGLEWMGYINPRSGYTHYNQKLKDKATLTADKSASTAYMELSSLRSEDTAVYYCA RSAYYDYDGFAYWGQGTLVTVSSGGGGSGGGGSGGGGSDIQMTQSPSSLSASVGDRVT ITCSASSSVSYMNWYQQKPGKAPKRLIYDTSKLASGVPSRFSGSGSGTDFTLTISSLQ PEDFATYYCQQWSSNPPTFGGGTKVEIKGGGGSGGGGSGGGGSQEDERIVLVDNKCKC ARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCD PTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTP DACYPDGGGGSGGGGSGGGGSDAHKSEVAHRFKDLGEENFKALVLIAFAQYLQQCPFE DHVKLVNEVTEFAKTCVADESAENCDKSLHTLFGDKLCTVATLRETYGEMADCCAKQE PERNECFLQHKDDNPNLPRLVRPEVDVMCTAFHDNEETFLKKYLYEIARRHPYFYAPE LLFFAKRYKAAFTECCQAADKAACLLPKLDELRDEGKASSAKQRLKCASLQKFGERAF KAWAVARLSQRFPKAEFAEVSKLVTDLTKVHTECCHGDLLECADDRADLAKYICENQD SISSKLKECCEKPLLEKSHCIAEVENDEMPADLPSLAADFVESKDVCKNYAEAKDVFL GMFLYEYARRHPDYSVVLLLRLAKTYETTLEKCCAAADPHECYAKVFDEFKPLVEEPQ NLIKQNCELFKQLGEYKFQNALLVRYTKKVPQVSTPTLVEVSRNLGKVGSKCCKHPEA KRMPCAEDYLSVVLNQLCVLHEKTPVSDRVTKCCTESLVNRRPCFSALEVDETYVPKE FNAETFTFHADICTLSEKERQIKKQTALVELVKHKPKATKEQLKAVMDDFAAFVEKCC KADDKETCFAEEGPKLVAASQAALGL
101	V15J15HSA(wt) DNA	ATGGGGTGGTCCTACATTATCCTGTTCCTCGTGGCCACCGCCACTGGCGTGCACTCAC AGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGGT GTCCTGCAAGGCCTCCGGCTACACCTTCATCAGCTACACCATGCACTGGGTGCGACAG GCCCCTGGACAGGGCCTGGAATGGATGGGCTACATCAACCCTAGATCTGGCTACACCC ACTACAACCAGAAGCTGAAGGACAAGGCCACCCTGACCGCCGACAAGTCTGCCTCCAC CGCCTACATGGAACTGTCCTCCCTGCGGAGCGAGGACACCGCCGTGTACTACTGTGCC AGATCCGCCTACTACGACTACGACGGCTTCGCCTATTGGGGCCAGGGCACCCTCGTGA CAGTGTCTAGCGGTGGCGGAGGATCTGGCGGAGGCGGTAGTGGCGGTGGCGGATCTGA TATCCAGATGACCCAGTCCCCCTCCAGCCTGTCTGCCTCTGTGGGCGACAGAGTGACA ATTACCTGCTCCGCCAGCTCCTCCGTGTCTTACATGAACTGGTATCAGCAGAAGCCCG GCAAGGCCCCCAAGCGGCTGATCTACGACACCTCCAAGCTGGCCTCTGGCGTGCCCTC CAGATTCTCCGGCTCTGGCTCTGGCACCGACTTTACCCTGACCATCAGCTCCCTGCAG CCCGAGGACTTCGCCACCTACTACTGCCAGCAGTGGTCCTCCAACCCTCCCACCTTTG GCGGAGGCACCAAGGTGGAAATCAAAGGCGGCGGAGGAAGCGGGGGAGGCGGTTCTGG GGGTGGTGGATCTCAGGAAGATGAGCGGATCGTGCTGGTGGACAACAAGTGCAAGTGC GCCCGGATCACCTCCCGGATCATCCGGTCCTCCGAGGATCCCAACGAGGACATCGTGG AACGGAACATCAGAATCATCGTGCCCCTGAACAACCGCGAGAACATCTCCGACCCCAC

SEQ ID NO:	Short Name	Sequence
		CAGCCCTCTGCGGACCAGATTCGTGTACCACCTGTCCGACCTGTGCAAGAAGTGCGAC CCTACCGAGGTGGAACTGGACAACCAGATCGTGACCGCCACCCAGTCCAACATCTGCG ACGAGGACTCCGCCACCGAGACATGCTACACCTACGACCGGAACAAGTGCTACACCGC CGTGGTGCCTCTGGTGTACGGCGGCGAGACAAAGATGGTGGAAACCGCCCTGACCCCC GACGCCTGCTATCCTGATGGAGGCGGAGGATCTGGTGGCGGTGGTTCTGGCGGAGGGG GCTCTGACGCCCACAAATCGGAGGTAGCGCACCGGTTCAAAGACTTGGGAGAAGAAAA CTTTAAGGCCCTTGTACTCATTGCGTTTGCGCAGTATTTGCAGCAGTGCCCATTCGAG GACCATGTCAAACTTGTCAACGAAGTGACAGAGTTTGCGAAAACTTGCGTCGCCGACG AATCCGCGGAGAACTGTGACAAGTCGCTGCATACGTTGTTCGGGGATAAGCTCTGTAC CGTAGCGACCTTGAGGGAAACTTACGGGGAAATGGCGGACTGTTGCGCTAAGCAGGAG CCGGAACGGAACGAGTGTTTCCTTCAGCATAAGGATGACAACCCCAACCTCCCTAGAT TGGTCAGACCCGAAGTGGATGTGATGTGCACAGCATTCCATGACAATGAGGAAACCTT TCTCAAAAAGTATTTGTACGAGATTGCCCGACGACACCCCTATTTCTACGCTCCCGAG TTGCTCTTCTTCGCGAAACGGTATAAAGCTGCCTTTACTGAATGCTGTCAAGCAGCGG ACAAGGCCGCATGCCTCCTTCCCAAATTGGATGAACTCCGCGATGAAGGGAAGGCGTC ATCGGCCAAACAGCGGCTTAAGTGCGCATCGCTTCAGAAATTCGGAGAGAGGGCGTTC AAAGCGTGGGCCGTCGCGAGACTGTCGCAGAGATTCCCTAAGGCGGAATTTGCAGAGG TATCGAAGCTCGTGACAGACCTCACAAAGGTCCACACCGAATGTTGCCATGGAGACCT GCTTGAGTGCGCCGATGATAGGGCAGACCTCGCAAAGTACATTTGTGAGAATCAGGAC AGCATTAGCTCCAAGCTGAAAGAGTGCTGTGAGAAGCCTTTGCTGGAAAAATCCCACT GTATCGCCGAGGTAGAAAACGATGAAATGCCCGCTGATCTTCCCTCGCTGGCGGCAGA CTTCGTCGAGTCGAAGGACGTCTGCAAGAATTACGCAGAGGCAAAAGATGTGTTTCTT GGAATGTTCCTTTATGAGTATGCGAGAAGGCACCCGGATTATTCCGTGGTACTGCTCT TGCGATTGGCGAAAACGTACGAAACAACGCTTGAGAAGTGTTGTGCGGCTGCCGACCC GCATGAGTGCTACGCCAAGGTATTTGATGAGTTTAAACCTCTTGTCGAGGAACCCCAG AATCTTATCAAGCAGAACTGCGAGCTTTTCAAGCAGTTGGGTGAATACAAATTCCAGA ACGCGCTTCTGGTGAGGTATACCAAGAAAGTACCTCAAGTCTCAACACCCACACTCGT CGAGGTGTCACGGAACCTCGGGAAAGTAGGGTCGAAGTGCTGTAAACACCCAGAGGCC AAGCGCATGCCCTGTGCGGAGGACTACCTCTCGGTAGTGTTGAATCAACTGTGTGTCC TCCACGAAAAGACGCCGGTGTCAGACCGCGTCACAAAGTGCTGCACGGAGAGCCTGGT CAATAGACGCCCCTGCTTCTCAGCGCTGGAGGTGGATGAGACATACGTCCCGAAAGAG TTTAACGCCGAAACGTTTACTTTTCATGCTGATATCTGTACGTTGTCAGAGAAGGAAA GGCAAATCAAGAAACAAACTGCGCTTGTGGAACTGGTGAAGCACAAACCGAAGGCGAC TAAGGAACAGCTGAAGGCGGTGATGGATGACTTTGCCGCGTTCGTAGAGAAATGCTGT AAAGCAGACGATAAGGAGACTTGTTTTGCGGAAGAGGGAAAGAAACTTGTTGCTGCAA GTCAAGCTGCCTTAGGCTTATAG
102	V15J15HSA(wt) AA	MGWSYIILFLVATATGVHSQVQLVQSGAEVKKPGASVKVSCKASGYTFISYTMHWVRQ APGQGLEWMGYINPRSGYTHYNQKLKDKATLTADKSASTAYMELSSLRSEDTAVYYCA RSAYYDYDGFAYWGQGTLVTVSSGGGGSGGGGSGGGGSDIQMTQSPSSLSASVGDRVT ITCSASSSVSYMNWYQQKPGKAPKRLIYDTSKLASGVPSRFSGSGSGTDFTLTISSLQ PEDFATYYCQQWSSNPPTFGGGTKVEIKGGGGSGGGGSGGGGSQEDERIVLVDNKCKC ARITSRIIRSSEDPNEDIVERNIRIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCD PTEVELDNQIVTATQSNICDEDSATETCYTYDRNKCYTAVVPLVYGGETKMVETALTP DACYPDGGGGSGGGGSGGGGSDAHKSEVAHRFKDLGEENFKALVLIAFAQYLQQCPFE DHVKLVNEVTEFAKTCVADESAENCDKSLHTLFGDKLCTVATLRETYGEMADCCAKQE PERNECFLQHKDDNPNLPRLVRPEVDVMCTAFHDNEETFLKKYLYEIARRHPYFYAPE LLFFAKRYKAAFTECCQAADKAACLLPKLDELRDEGKASSAKQRLKCASLQKFGERAF KAWAVARLSQRFPKAEFAEVSKLVTDLTKVHTECCHGDLLECADDRADLAKYICENQD SISSKLKECCEKPLLEKSHCIAEVENDEMPADLPSLAADFVESKDVCKNYAEAKDVFL GMFLYEYARRHPDYSVVLLLRLAKTYETTLEKCCAAADPHECYAKVFDEFKPLVEEPQ NLIKQNCELFKQLGEYKFQNALLVRYTKKVPQVSTPTLVEVSRNLGKVGSKCCKHPEA KRMPCAEDYLSVVLNQLCVLHEKTPVSDRVTKCCTESLVNRRPCFSALEVDETYVPKE FNAETFTFHADICTLSEKERQIKKQTALVELVKHKPKATKEQLKAVMDDFAAFVEKCC KADDKETCFAEEGKKLVAASQAALGL

Patentkrav

1. Bindingsmolekyle, der omfatter et IgM-, IgA-, IgG/IgM- eller IgG/IgA-antistof med en modificeret J-kæde, hvor IgG/IgM- eller IgG/IgA-antistoffet indeholder et IgM- eller IgA-endestykke i IgG-tungkæden, hvor den modificerede J-kæde omfatter en absorptions-fordelings-metabolisme-udskillelses-(ADME)-modulerende del, der reducerer clearance af antistoffet fra et individs blodomløb, hvor den ADME-modulerende del er lokaliseret i den C-terminale ende eller den N-terminale ende af den modificerede J-kæde, og hvor den ADME-modulerende del omfatter et albuminprotein, et albuminbindende peptid, et albuminbindende antistoffragment, et FcRn-bindende peptid eller et FcRn-bindende antistoffragment.

2. Bindingsmolekyle ifølge krav 1, hvor den ADME-modulerende del omfatter humant serumalbumin.

3. Bindingsmolekyle, der omfatter et IgM-, IgA-, IgG/IgM- eller IgG/IgA-antistof med en modificeret J-kæde, hvor IgG/IgM- eller IgG/IgA-antistoffet indeholder et IgM- eller IgA-endestykke i IgG-tungkæden, hvor den modificerede J-kæde omfatter en ADME-modulerende del, der forøger en koncentration af bindingsmolekylet i et væv fra centralnervesystemet hos et individ, hvor den ADME-modulerende del er lokaliseret i den C-terminale ende eller den N-terminale ende af den modificerede J-kæde, og hvor den ADME-modulerende del omfatter et transferrinprotein, et leptinprotein, et transferrinreceptorbindende antistoffragment, et transferrinbindende antistoffragment, et insulinreceptorbindende antistoffragment, et IGF-1-receptorbindende antistoffragment eller et leptinreceptorbindende antistoffragment.

4. Bindingsmolekyle ifølge krav 3, hvor den ADME-modulerende del omfatter et transferrinprotein eller et leptinprotein.

5. Bindingsmolekyle, der omfatter et IgM-, IgA-, IgG/IgM- eller IgG/IgA-antistof med en modificeret J-kæde, hvor IgG/IgM- eller IgG/IgA-antistoffet indeholder et IgM- eller IgA-endestykke i IgG-tungkæden, hvor den modificerede J-kæde omfatter en ADME-modulerende del, der forøger retention af bindingsmolekylet i et ekstravaskulært rum hos et individ, hvor den ADME-modulerende del er lokaliseret i den C-terminale ende eller den N-terminale ende af den modificerede J-kæde, og hvor den ADME-modulerende del omfatter et hyaluronsyrebindende protein (HABP), et hyaluronsyrebindende antistoffragment, et TSG-6-protein eller et TSG-6-bindende antistoffragment.

6. Bindingsmolekyle ifølge et hvilket som helst af kravene 1 til 2, hvor IgM-, IgA-, IgG/IgM- eller IgG/IgA-antistoffet binder til et hæmatologisk cancermål, der er CD20.

7. Bindingsmolekyle ifølge et hvilket som helst af kravene 1 til 5, hvor IgM-, IgA-, IgG/IgM- eller IgG/IgA-antistoffet binder til beta-sekretase 1 (BACE).

8. Bindingsmolekyle ifølge et hvilket som helst af de foregående krav, hvor den modificerede J-kæde omfatter den native humane J-kædesekvens ifølge SEQ ID NO: 1.

9. Bindingsmolekyle ifølge krav 8, hvor den ADME-modulerende del indføres i den native humane J-kædesekvens ifølge SEQ ID NO: 1 ved direkte eller indirekte fusion, og hvor indirekte fusion sker ved hjælp af en peptidlinker.

10. Bindingsmolekyle ifølge krav 8, hvor den ADME-modulerende del indføres i den native humane J-kædesekvens ifølge SEQ ID NO: 1 ved kemisk eller kemoenzymatisk derivatisering.

11. Bindingsmolekyle ifølge krav 10, hvor den ADME-modulerende del indføres i den native humane J-kæde med en spaltelig eller ikke-spaltelig linker, hvor den spaltelige linker er en kemisk labil linker eller en enzymlabil linker.

12. Bindingsmolekyle ifølge krav 11, hvor linkeren er valgt fra gruppen bestående af N-succinimidyl-3-(2-pyridyldithio)propionat (SPDP), succinimidyl-4-(N-maleimidomethyl)cyclohexan-1-carboxylat (SMCC), N-succinimidyl-4-(2-pyridylthio)pentanoat (SPP), iminothiolan (IT), bifunktionelle derivater af imidoestere, aktive estere, aldehyder, bis-azidoforbindelser, bis-diazoniumderivater, diisocyanater og bis-aktive fluorforbindelser.

13. Bindingsmolekyle ifølge et hvilket som helst af kravene 1-12 til anvendelse ved behandling af cancer.

14. Bindingsmolekyle til anvendelse ifølge krav 13, hvor canceren er en hæmatologisk cancer, en epitelcancer eller en cancer i centralnervesystemet.

DRAWINGS

Drawing

FIG. 1

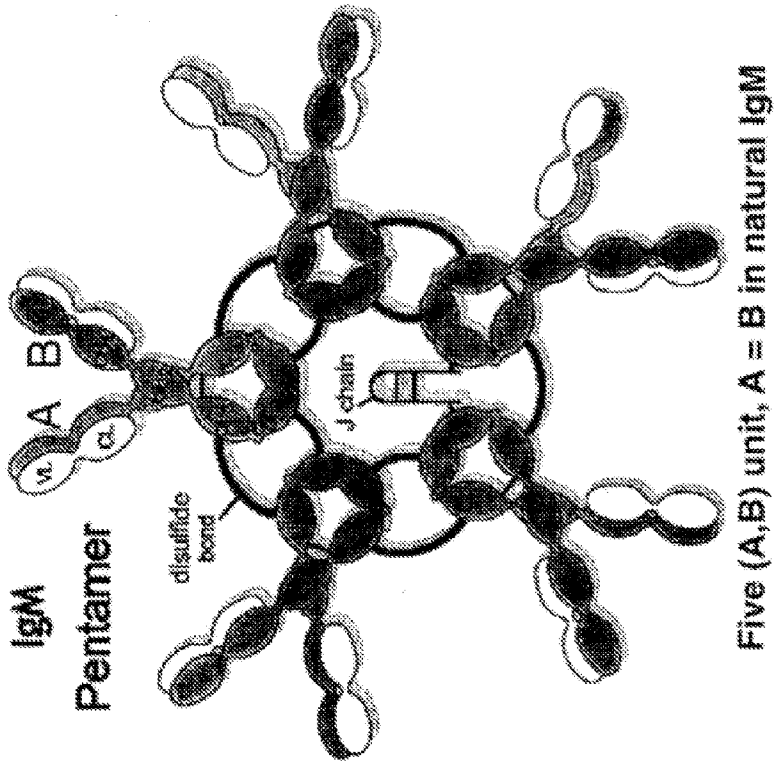


FIG. 2

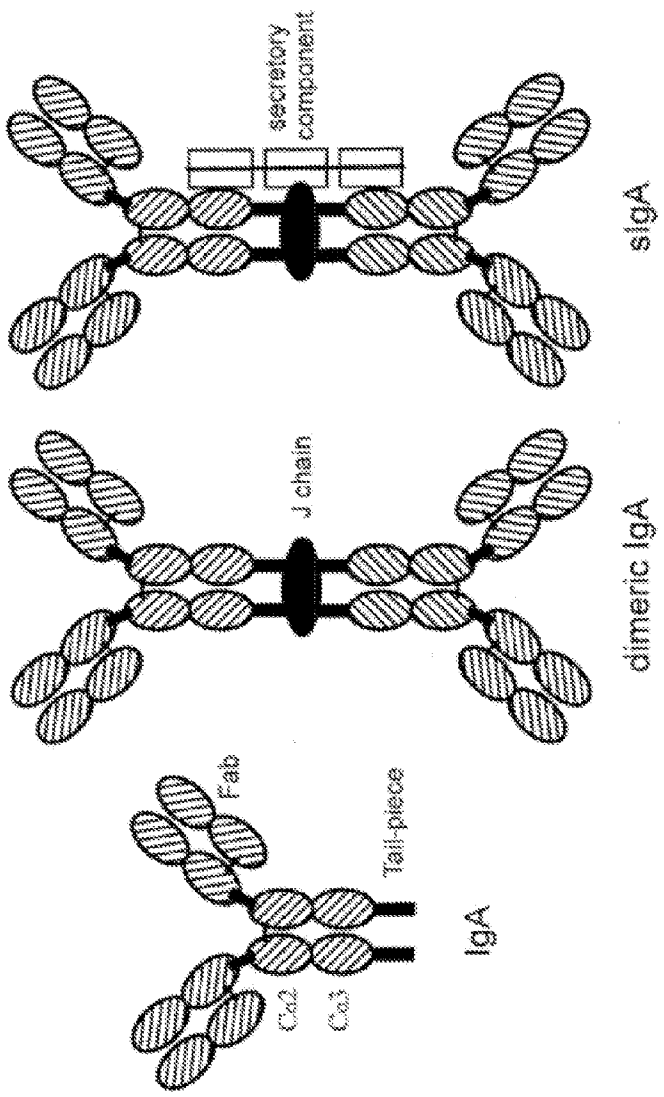

secretory component
sIgA
J chain
dimeric IgA
Fab
Tail-piece
IgA
Cα2
Cα3

FIG. 3

Mature Human J Chain

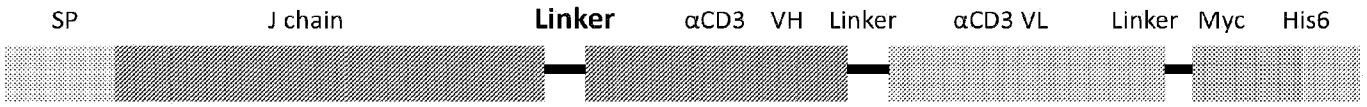
- QEDERIVLVDNKCKCARITSRIIRSSEDPNEDIVERNI RIIVPLNNRENISDPTSPLRTRFVYHLSDLCKKCDPT EVELDNQIVTATQSNICDEDSATETCYTYDRNKCYT AVVPLVYGGETKMVETALTPDACYPD (SEQ ID NO: 1)
- Number of amino acids: 137
- Molecular weight: 15594.4
- Theoretical pI: 4.59

DK/EP 3355913 T3

FIG. 4A
Orientation of J-chain Constructs

J-CD3$_{scFv}$:

J-chain at N-terminus scFv anti-CD3 at C-terminus



CD3$_{scFv}$-J:

scFv anti-CD3 at N-terminus **J-chain** at C-terminus

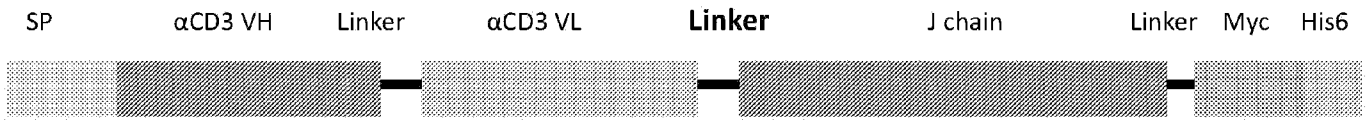


FIG. 4B
Orientation of J-chain Constructs

J-HSA:

J-chain at N-terminus HSA at C-terminus

HSA-J:

HSA at N-terminus **J-chain** at C-terminus

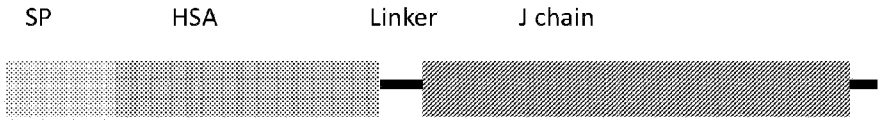


FIG. 5

Asymmetric IgM Pentamer with modified J-chain

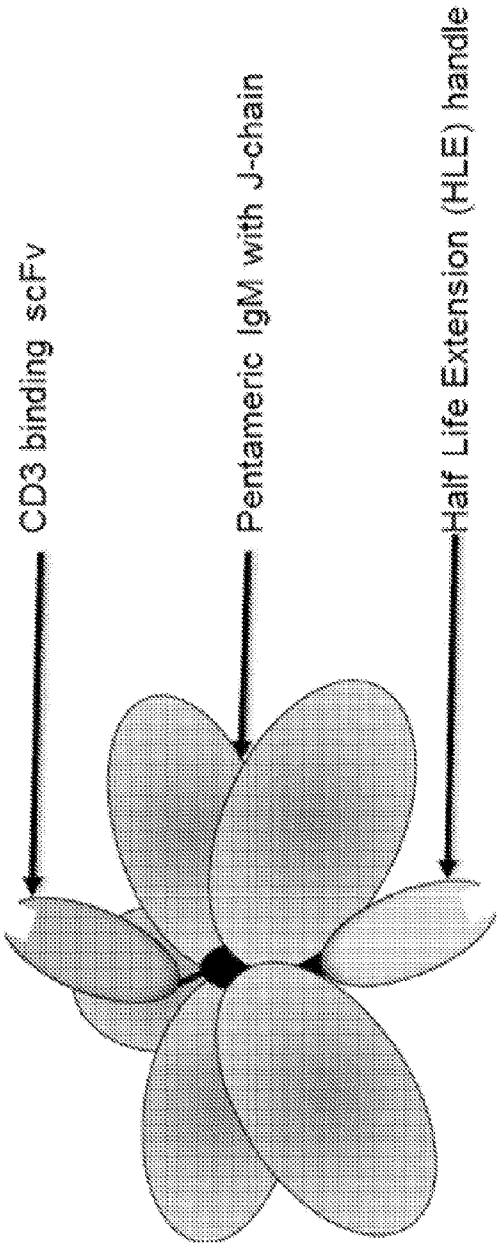


FIG. 6

Anti-CD20 $_{IgM}$ x Anti-CD3$_J$ bi-specific IgM can be expressed and assembled into pentamer with various scFv at either N- or C- terminus of J-chain

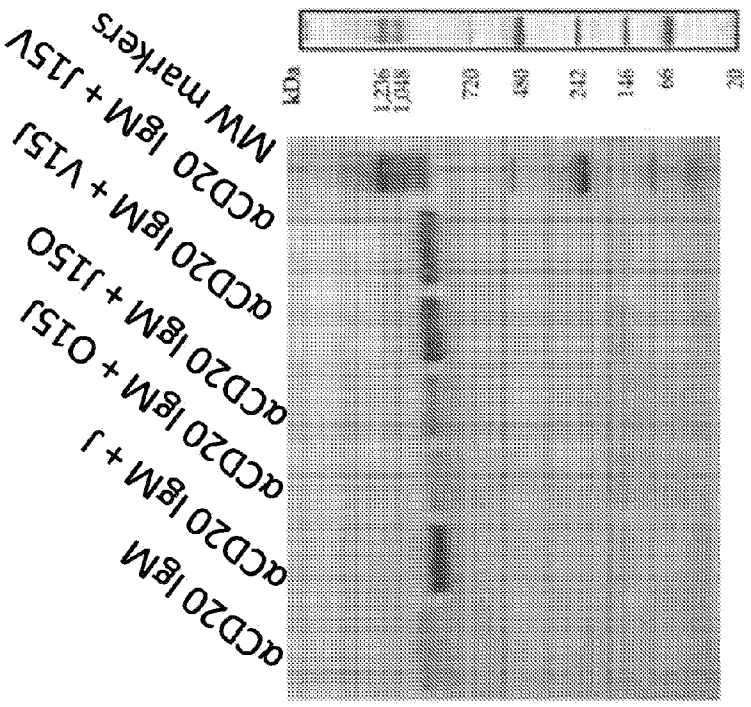
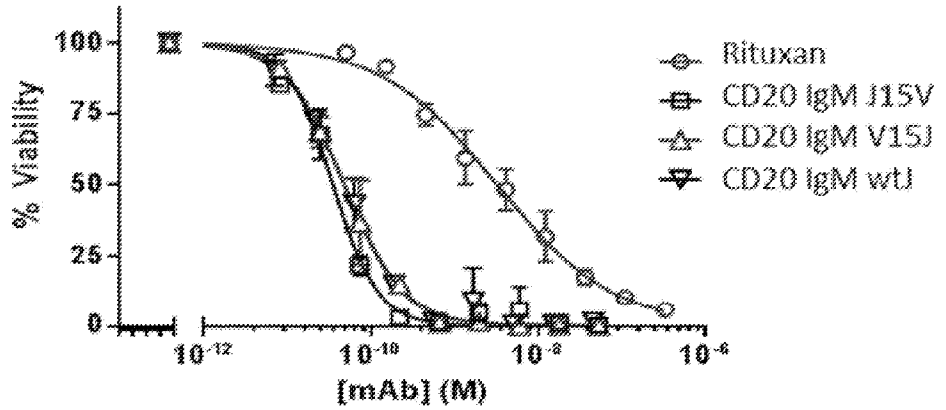


FIG. 7

Anti-CD20 $_{IgM}$ x Anti-CD3$_J$ bi-specific IgM with Anti-CD3 scFv at N- or C- terminus of J-chain: Bi-specific IgMs are functional in CDC



EC50 (nM)			
Rituxan	CD20 IgM J15V	CD20 IgM V15J	CD20 IgM wtJ
3.29	0.04	0.05	0.05

FIG. 8

Anti-CD20 $_{IgM}$ x Anti-CD3$_J$ Bi-specific IgM
Induces T-cell activation in presence of target cells

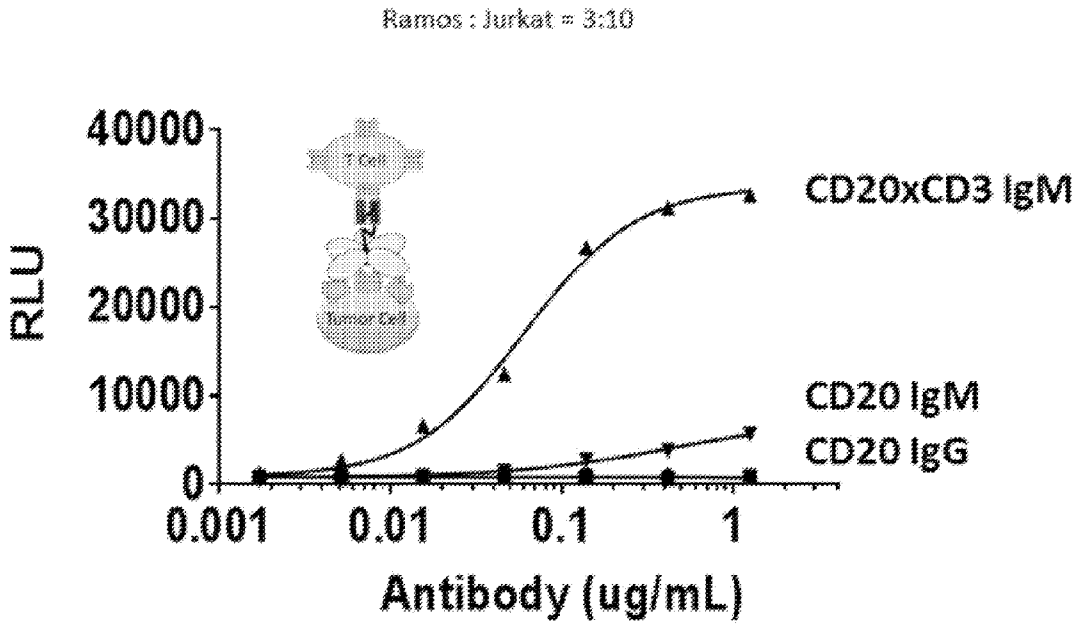
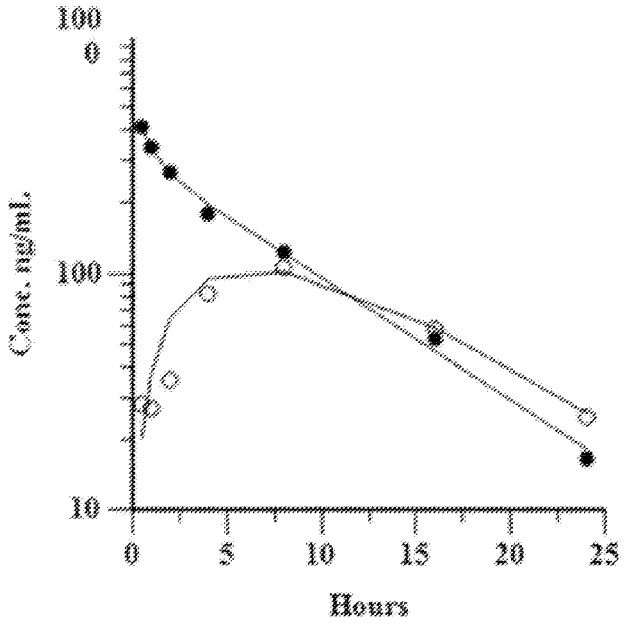


FIG. 9

IGM-55.5 has reduced in-vivo half life in mice in the absence of half-life extension

Panel A



Panel B

Mean (± SD) Plasma Pharmacokinetic Parameters for IGM 55.5 in Mice (N = 3)

Route	IV	IP
CL (mL/hr/kg)	7.1	
Vss (mL/kg)	56	
Cmax (µg/mL)	563	106
T1/2α (hr)	0.49	
T1/2β (hr)	5.83	
AUMC (µg hr hr/mL)	22,400	1540
MRT (hr)	7.94	

FIG. 10
Multimer specific ELISA for CD20 IgMs

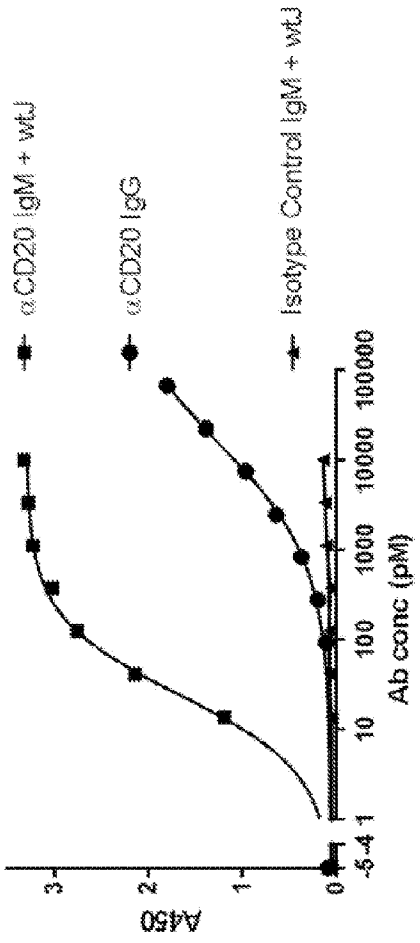
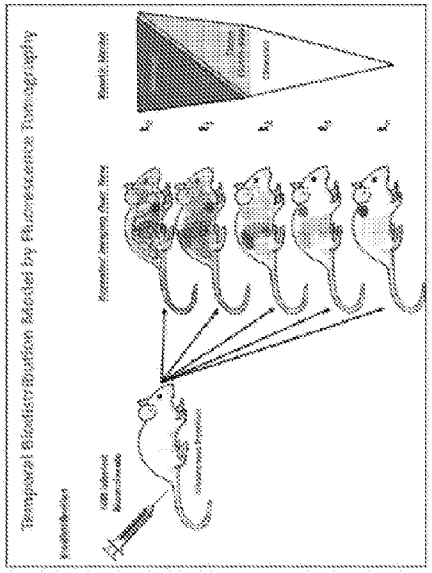


FIG. 11

Panel A



Panel B

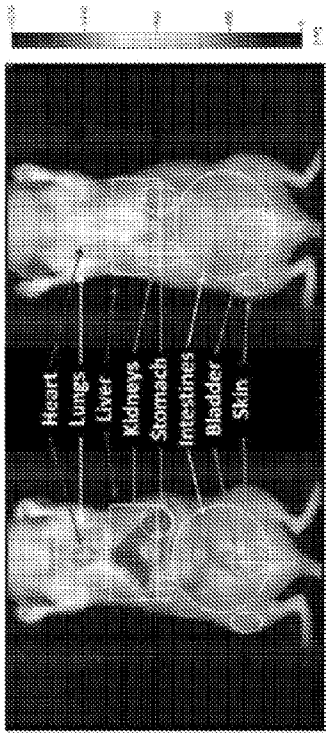
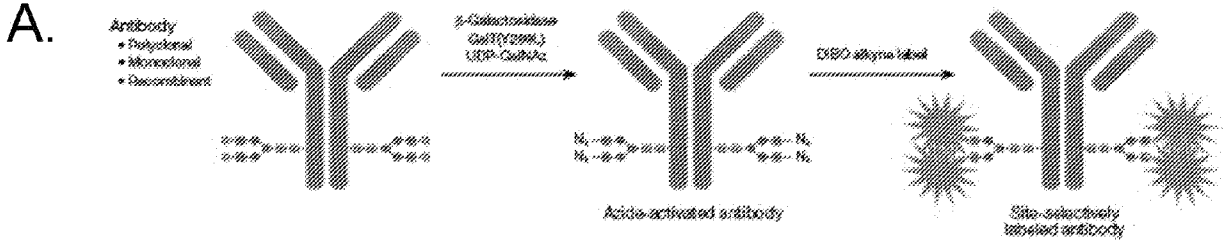
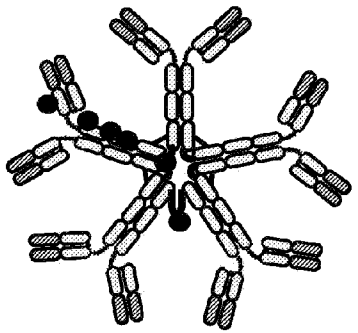


FIG. 12

Site Specific labeling of IgM glycans using chemo-enzymatic approaches





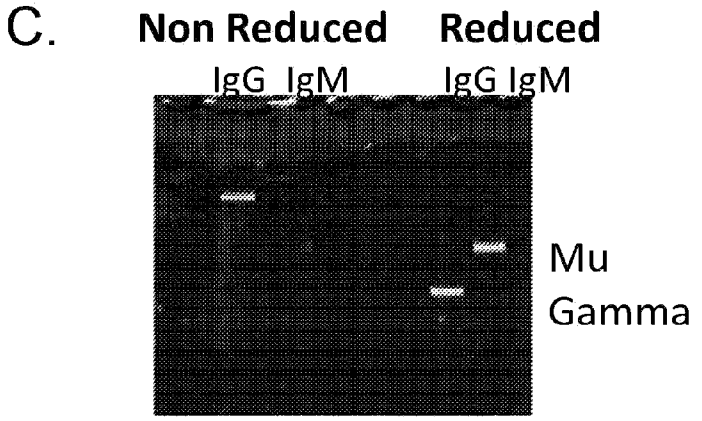


FIG. 13

Combinatorial approach to compartmentalization uses of J-chain binding molecules:

Antibody Target	J-chain binding moiety target
Super agonist targets: CD137 (4-1BB), OX40, CD40, GITR, CD27, HVEM **Low expression level targets:** EGFR, HER2, HER3, EpCAM, CEACAM, Gp100, MAGE1 **Low affinity targets:** NY-ESO-1, Sialyl Lewis X antigen, Tn antigen **Hematologic cancer targets:** CD19, CD20, CD22, CD33, CD38, CD52, CD70 **Other binding targets:** VEGF, TNF alpha, amyloid beta, BACE	**Targets for regulating half-life** Human serum albumin (HSA) HSA binding peptides Neonatal Fc Receptor (FcRn) Fc domain **Targets for regulating bio-distribution** Transferrin, Transferrin receptor (TfR) Insulin, Insulin Receptor IGF-1, IGF-1 receptor Leptin, Leptin receptor basigin Glut1 CD98hc **Target for retention in intra-ocular or intra-articular compartments** Hyaluronic Acid TSG-6

DK/EP 3355913 T3

FIG. 14

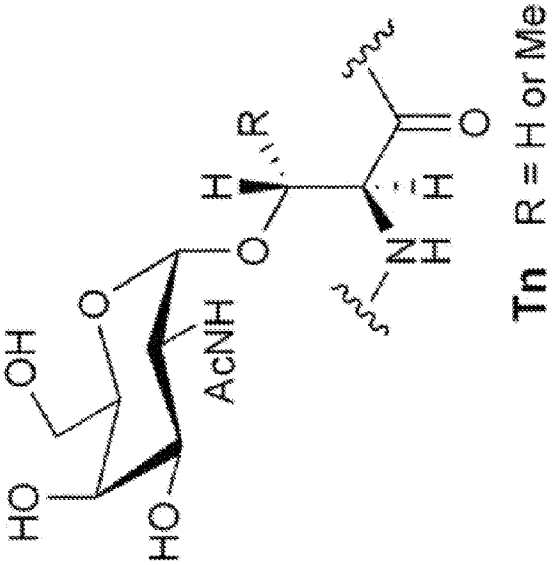
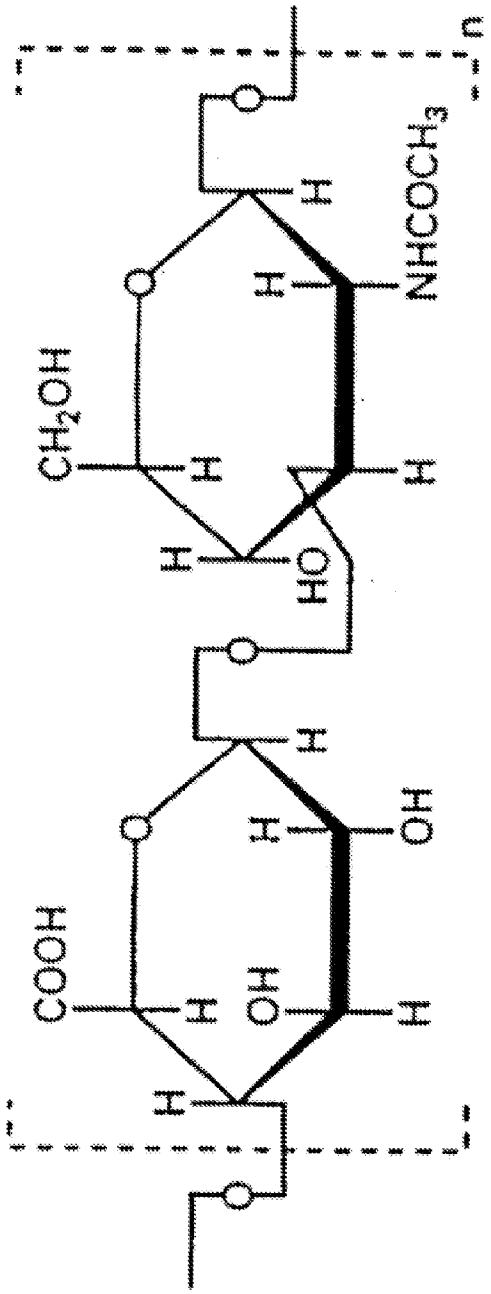


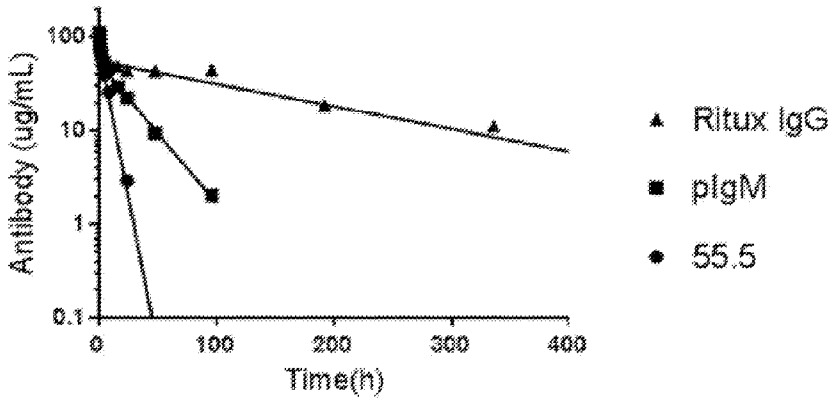
FIG. 15



Alternating units of 1,4-linked
N-acetylglucosamine and glucuronic acid

FIG. 16

Panel A:

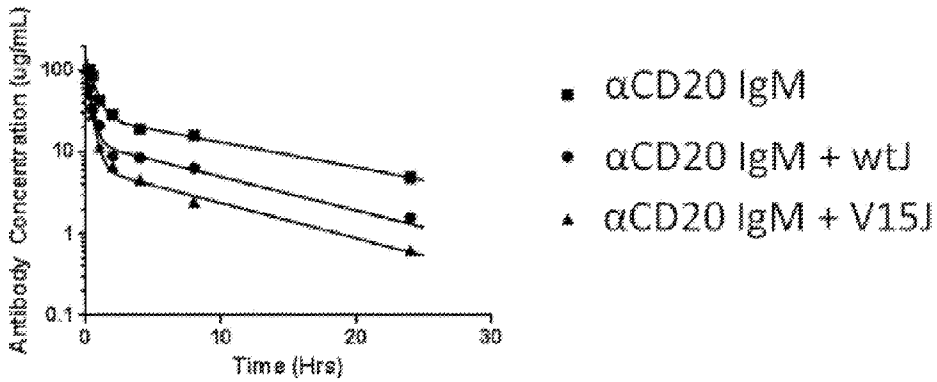


Panel B:

Antibody	$t_{1/2}$ α (hrs)	$t_{1/2}$ β (hrs)	$AUC_{0-\infty}$ (µg/ml*hr)
Rituximab	2.4	158.7	12180
Polyclonal IgM	0.99	14.3	1412
55.5 (CHO IgM)	0.2	4.7	549

FIG. 17

Panel A:

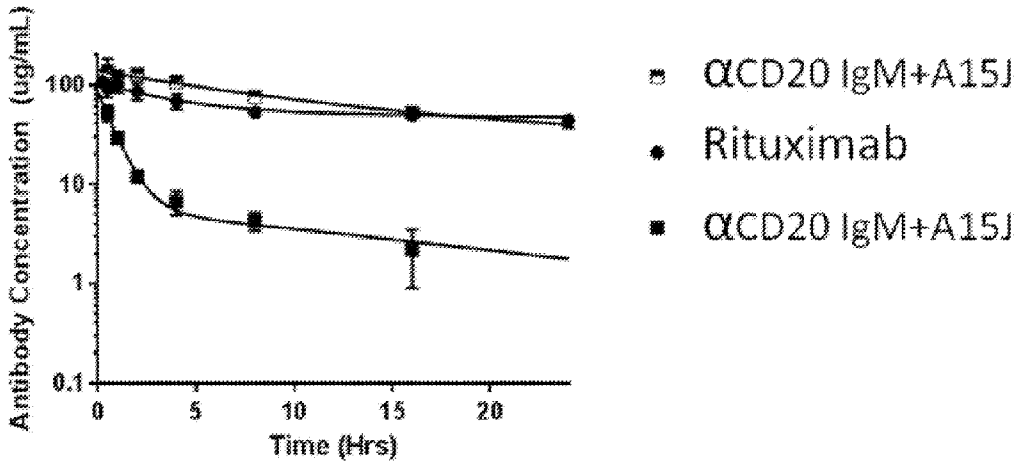


Panel B:

Antibody	$t_{1/2}$ α (hrs)	$t_{1/2}$ β (hrs)	$AUC_{0-\infty}$ (µg/ml*hr)
αCD20 IgM	0.38	9.8	477
αCD20 IgM + wtJ	0.24	7.3	169
αCD20 IgM + V15J	0.23	4.0	82

FIG. 18

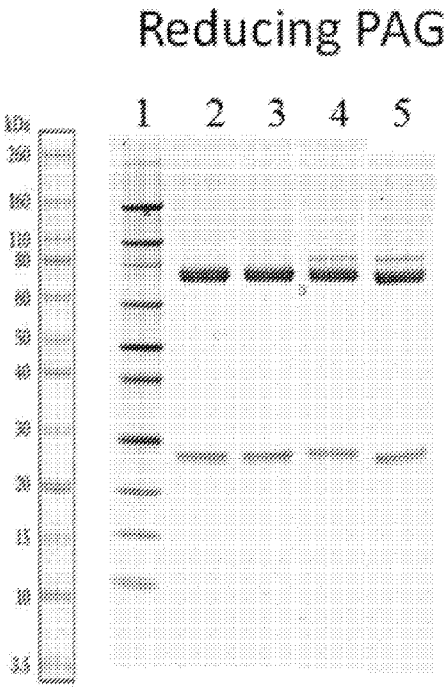
Panel A:

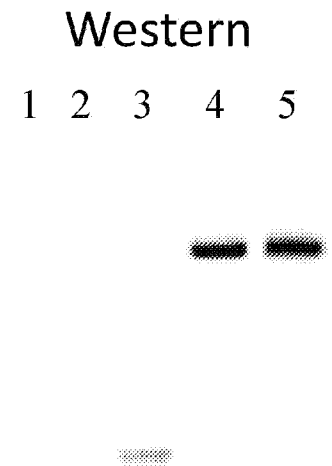


Panel B:

Antibody	$t_{1/2}$ α (hrs)	$t_{1/2}$ β (hrs)	$AUC_{0-\infty}$ (μg/ml*hr)
Rituximab	2.4	158.7	12180
αCD20 IgM+V15J	0.23	4.0	82
αCD20 IgM+A15J	3.2	32.4	1341

FIG. 19





*All samples loaded at 1 ug/well.

Sample
1 LMW Marker
2 αCD20 IgM
3 αCD20 IgM +J
4 αCD20 IgM+ HSA-J
5 αCD20 IgM+ J-HSA

FIG. 20

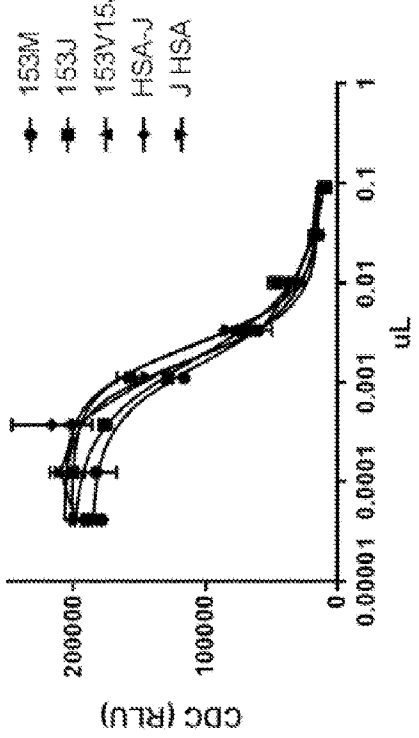

153M
153J
153V15J
HSA-J
J HSA
CDC (RLU)
200000
100000
0
0.00001
0.0001
0.001
0.01
0.1
1
uL

FIG. 21

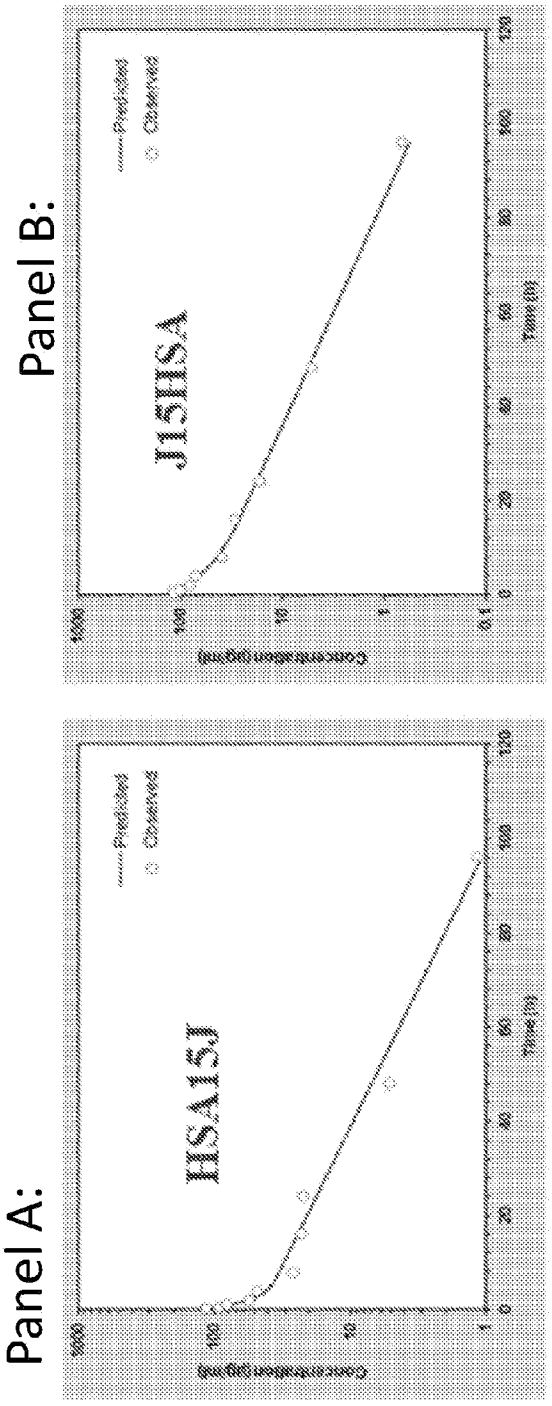
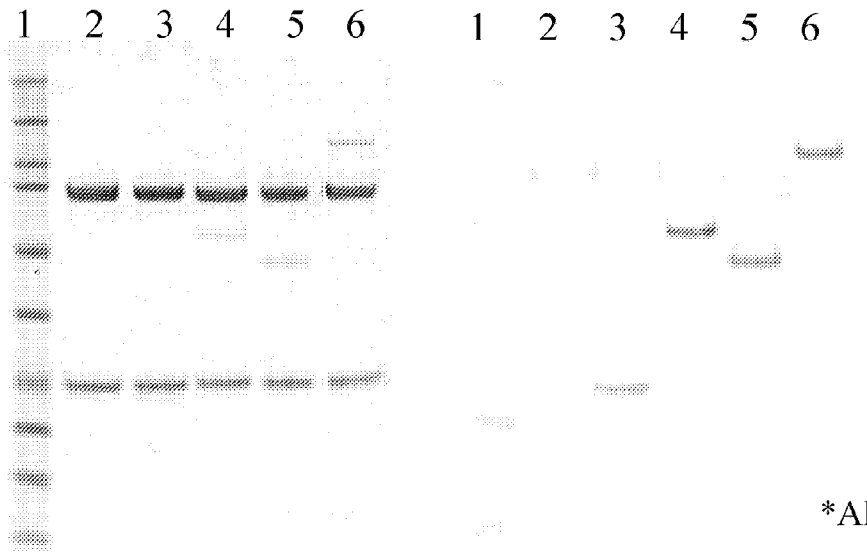

Panel A:
HSA15J
Predicted
Observed
Concentration (ng/mL)
Time (h)
Panel B:
J15HSA
Predicted
Observed
Concentration (ng/mL)
Time (h)

FIG. 22

Antibody	$t_{1/2}$ α (hrs)	$t_{1/2}$ β (hrs)	$AUC_{0-\infty}$ (μg/ml* hr)
Rituximab	2.4	158.7	12180
1.5.3 IgM+V15J	0.23	4.0	82
1.5.3 IgM+J15A	3.2	32.4	1341
1.5.3 IgM+A15J	0.85	10.3	1196
1.5.3 IgM+J15H	2.3	14.5	1380
1.5.3 IgM+H15J	0.71	17.7	1259

FIG. 23



*All samples loaded at 1.2 ug/ml

Reducing PAGE Western

1. Marker
2. 1.5.3 IgM
3. 1.5.3 IgM + wt J
4. 1.5.3 + V15J15ABD
5. 1.5.3+V15J
6. 1.5.3+V15J15HSA

FIG. 24

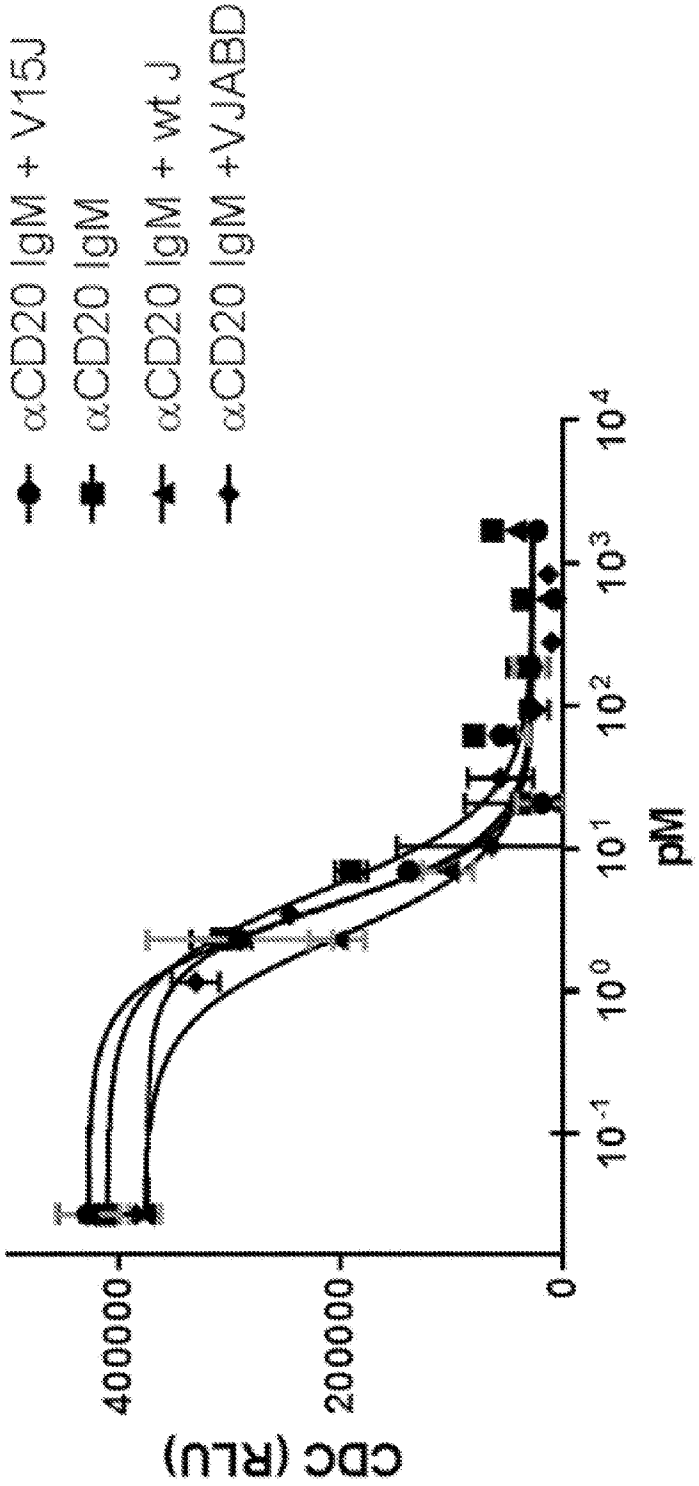

αCD20 IgM + V15J
αCD20 IgM
αCD20 IgM + wt J
αCD20 IgM +VJABD
CDC (RLU)
400000
200000
0
10-1
100
101
102
103
104
pM

FIG. 25

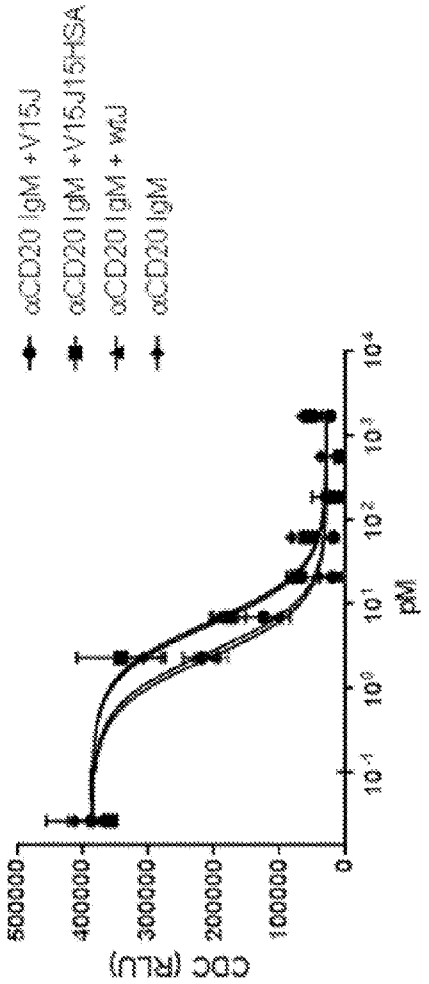

αCD20 IgM +V15J
αCD20 IgM +V15J15-HSA
αCD20 IgM + wtJ
αCD20 IgM
CDC (RLU)
500000
400000
300000
200000
100000
0
10⁻¹
10⁰
10¹
10²
10³
10⁴
pM

FIG. 26

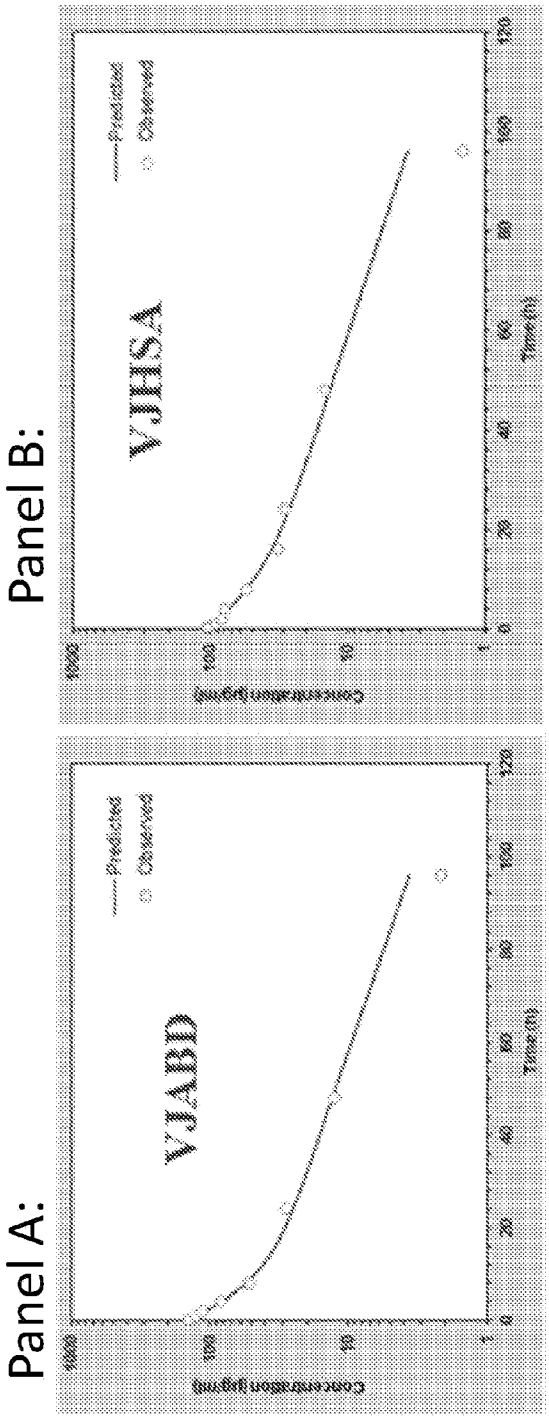
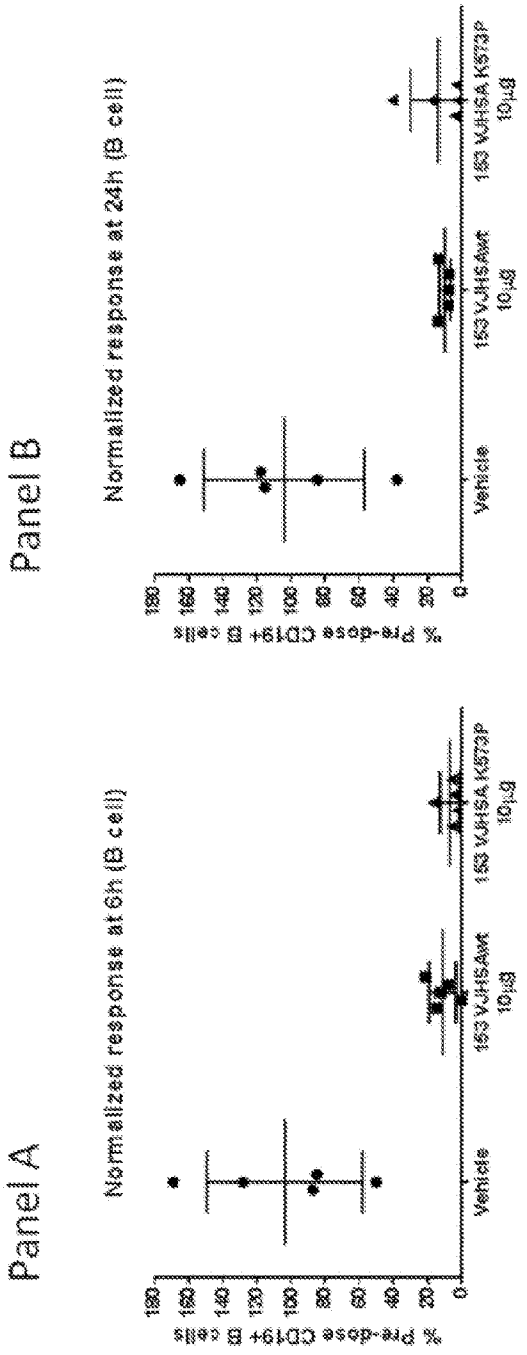

Panel A:
VJABD
Predicted
Observed
Concentration (µg/mL)
Time (h)
Panel B:
VJHSA
Predicted
Observed
Concentration (µg/mL)
Time (h)

FIG. 27

	$t_{1/2\ Alpha}$	$t_{1/2\ Beta}$	AUC_{0-inf}
153 IgM HSAJ	0.78	17.8	1260
153 IgM JHSA	2.33	14.5	1380
153 IgM VJABD	3.22	26.0	2212
153 IgM VJHSA	4.22	25.6	2167

FIG. 28


Panel A
Normalized response at 6h (B cell)
% Pre-dose CD19+ B cells
160
140
120
100
80
60
40
20
0
Vehicle
153 VJH5Amt 10µg
153 VJH5A K573P 10µg
Panel B
Normalized response at 24h (B cell)
% Pre-dose CD19+ B cells
160
140
120
100
80
60
40
20
0
Vehicle
153 VJH5Amt 10µg
153 VJH5A K573P 10µg