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(54) Title: ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

(57) Abstract: The applications provides the anti-CD3 monoclonal antibodies, antigen-binding portions thereof, therapeutic compositions thereof and/or nucleic acid encoding the same, and their use to active CD3+ T-cells to enhance cell-mediated immune responses in the treatment of cancer and other T-cell dysfunctional disorders.

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of U.S. Provisional Patent Application No. 62551032 filed 27-AUG-2017, the entire disclosure of which is expressly incorporated herein by reference in its entirety.

TECHNICAL FIELD

[0002] The present disclosure generally relates to the technical field of antibodies, and more particularly relates to making and using anti-CD3 antibodies.

BACKGROUND

[0003] Cancer is a major health problem across the world. In the United States alone it is estimated that in 2016 there were 1,685,210 new cases of cancer diagnosed and 595,690 deaths from the disease (http://www.cancer.gov). As such, any pharmaceutical agent that can reduce the severity or mortality rate from cancer is desirable.

[0004] In the immune system, resting T-cells can be activated to respond to antigen through a primary signal delivered through the T-cell receptor (TCR) by foreign antigen peptides presented by antigen-presenting cells (APCs). In addition to this primary signal, there are secondary positive and negative co-stimulatory signals that further influence the response of the T-cells. A secondary positive signal is required for full T-cell activation ((Lafferty et al., Ausl. J. Exp. Biol. Med. Sci. 53: 27-42 (1975)). Negative secondary signals can result in T-cell suppression and tolerance.

[0005] The Cluster of Differentiation 3 (CD3) T-cell co-receptor helps to activate both the cytotoxic T-Cell (CD8+ naive T cells) and also T helper cells (CD4+ naive T cells). It consists of a protein complex and is composed of four distinct chains. In mammals, the complex contains a CD3γ chain, a CD3δ chain, and two CD3ε chains. These chains associate with a molecule known as the T-cell receptor (TCR) and the ζ-chain (zeta-chain) to generate an activation signal in T lymphocytes. The TCR, ζ-chain, and CD3 molecules together constitute the TCR complex.

[0006] Because CD3 is required for T-cell activation, monoclonal antibodies (mAbs) that target it are being investigated as immunosuppressant therapies (e.g., otelixizumab, foralumab) for type 1 diabetes and other autoimmune diseases. However, it remains unclear whether specific anti-CD3 mAbs can active CD3+ T cells, thus to increase the host immune response to cancerous tumours.

SUMMARY

[0007] The present disclosure provides, among others, anti-CD3 monoclonal antibodies, antigen-binding portions thereof, therapeutic compositions thereof and/or nucleic acid encoding the same, and their use to activate CD3+ T-cells and thus to enhance cell-mediated immune responses in the treatment of cancer and other T-cell dysfunctional disorders.

[0008] In one embodiment, an isolated monoclonal antibody (mAb) or antigen-binding fragment thereof that binds specifically to human CD3 is provided.

[0009] In one embodiment, the isolated mAb or antigen-binding fragment comprises an amino acid sequence having a percentage homology with SEQ ID NO:4, SEQ ID NO:8, SEQ ID NO:12, SEQ ID NO:16, SEQ ID NO:20, SEQ ID NO:24, SEQ ID NO:28, SEQ ID NO:32, SEQ ID NO:36, SEQ ID NO:40, SEQ ID NO:44, SEQ ID NO:48, SEQ ID NO:52, SEQ ID NO:56, SEQ ID NO:60, SEQ ID NO:64, SEQ ID NO:68, or SEQ ID NO:72. In one embodiment, the percentage homology is not less than 70%, 75%, 80%, 90%, 95%, 98%, or 99%.

[0010] In one embodiment, the isolated mAb or antigen-binding fragment has a binding affinity to CD3 with a Kd not greater than 30nM, 40nM or 50nM. In one embodiment, the isolated mAb or antigen-binding fragment has a binding affinity to CD3 with a Kd not greater than 60nM. In one embodiment, the isolated mAb or antigen-binding fragment has a binding affinity to CD3 with a Kd not greater than 70nM. In one embodiment, the isolated mAb or antigen-binding fragment has a binding affinity to CD3 with a Kd not greater than 80nM. In one embodiment, the isolated mAb or antigen-binding fragment has a binding affinity to CD3 with a Kd not greater than 100nM.

[0011] In some embodiments, the isolated mAb or antigen-binding fragment may exhibit one or more functional properties including, for example, high affinity binding to CD3, enhancing T cell activation, stimulating antibody response, reversing the suppressive function of an immunosuppressive cell, or a combination thereof. In one embodiment, the isolated mAb or antigen-binding fragment may enhance T-cell activation via T-cell proliferation, IFN-γ and/or IL-2 secretion, or a combination thereof. In one embodiment, the isolated mAb or antigen-binding fragment may reverse the suppressive function of a T regulatory cell.

[0012] In some embodiments, the isolated mAb or antigen-binding fragment may include a human framework region. In one embodiment, the isolated mAb or antigen-binding fragment may include a humanized antibody, a chimeric antibody, or a recombinant antibody.

[0013] In one embodiment, the isolated mAb or antigen-binding fragment may be an antibody belonging to IgG family. In one embodiment, the isolated mAb is an IgG. In one embodiment, the isolated mAb or antigen-binding fragment may include an antigen-binding fragment including, for example, a Fv, a Fab, a F(ab')2, a scFV or a scFV2 fragment.

[0014] In one embodiment, the isolated mAb or antigen-binding fragment may be a bispecific antibody, tri-specific antibody, or multi-specific antibody.

[0015] In one embodiment, the isolated mAb or antigen-binding fragment may include an IgG1 heavy chain that comprises an amino acid sequence having a percentage homology with SEQ ID NO:7, SEQ ID NO:15, SEQ ID NO:23, SEQ ID NO:31, SEQ ID NO:39, SEQ ID NO:47, SEQ ID NO:55, SEQ ID NO:63, or SEQ ID NO:71. In one embodiment, the percentage homology is not less than 70%, 75%, 80%, 90%, 95%, 98%, or 99%.

[0016] In one embodiment, the isolated mAb or antigen-binding fragment may include a kappa light chain that comprises an amino acid sequence having a percentage homology with SEQ ID NO:3, SEQ ID NO:11, SEQ ID NO:19, SEQ ID NO:27, SEQ ID NO:35, SEQ ID NO:43, SEQ ID NO:51, SEQ ID NO:59, or SEQ ID NO:67. In one embodiment, the percentage homology is not less than 70%, 75%, 80%, 90%, 95%, 98%, or 99%.

[0017] In one embodiment, the isolated mAb or antigen-binding fragment may include a variable light chain that comprises an amino acid sequence having at least 70%, 80%, 90%, 95%, 98% or 99% identity with SEQ ID NO:4, SEQ ID NO:12, SEQ ID NO:20, SEQ ID NO:28, SEQ ID NO:36, SEQ ID NO:44, SEQ ID NO:52, SEQ ID NO:60, or SEQ ID NO: 68.

[0018] In one embodiment, the isolated mAb or antigen-binding fragment may include a variable heavy chain that comprises an amino acid sequence having a percentage homology with SEQ ID NO:8, SEQ ID NO:16, SEQ ID NO:24, SEQ ID NO:32, SEQ ID NO:40, SEQ ID NO:48, SEQ ID NO:56, SEQ ID NO:64, or SEQ ID NO:72. In one embodiment, the percentage homology is not less than 70%, 75%, 80%, 90%, 95%, 98%, or 99%.

[0019] The application further provides the nucleic acid sequences encoding the amino acid sequences, isolated mAbs or antigen-binding fragments disclosed herein. In one embodiment, the isolated nucleic acids encoding an IgG1 heavy chain having a percentage homology with SEQ ID NO:5, SEQ ID NO:13, SEQ ID NO:21, SEQ ID NO:29, SEQ ID NO:37, SEQ ID NO:45, SEQ ID NO:53, SEQ ID NO:61, or SEQ ID NO:69; the kappa light chain comprising SEQ ID NO:1, SEQ ID NO:9, SEQ ID NO:17, SEQ ID NO:25, SEQ ID NO:33, SEQ ID NO:41, SEQ ID NO:49, SEQ ID NO:57, or SEQ ID NO:65. In one embodiment, the isolated nucleic acids encoding an variable light chain having a percentage homology with SEQ ID NO:2, SEQ ID NO:10, SEQ ID NO:18, SEQ ID NO:26, SEQ ID NO:34, SEQ ID NO:42, SEQ ID NO:50, SEQ ID NO:58, or SEQ ID NO:66. In one embodiment, the isolated nucleic acids encoding a variable heavy chain having a percentage homology with SEQ ID NO:6, SEQ ID NO:14, SEQ ID NO:22, SEQ ID NO:30, SEQ ID NO:38, SEQ ID NO:46, SEQ ID NO:54, SEQ ID NO:62, or SEQ ID NO:70. In one embodiment, the percentage homology is not less than 70%, 75%, 80%, 90%, 95%, 98%, or 99%.

[0020] The application further provides expression vectors containing the nucleic acid sequences disclosed herein. In one embodiment, the vector is expressible in a cell. In one embodiment, an expression vector encoding an isolated mAb or antigen-binding fragment as disclosed herein.

[0021] In one embodiment, the expression vector described herein may be present in a cell and expressible. In one embodiment, the expression vector described herein may be present in a host cell and expressible. In one embodiment, host cells comprising the expression vector are provided, wherein the expression vector comprises a nucleic acid disclosed herein. The host cell can be a prokaryotic cell or a eukaryotic cell.

[0022] Methods of producing an antibody as disclosed herein are provided. In one embodiment, the method includes the steps of providing a host that contains an expression vector expressible in the host cell, the expression vector comprises a nucleic acid sequence disclosed herein, to produce an antibody by the expression of the nucleic acid sequence.

[0023] The application further provides immunoconjugates. In one embodiment, the immunoconjudates include the isolated mAb or an antigen-binding fragment thereof conjugated to a drug unit. In some embodiments, the drug unit is linked to the isolated mAb or an antigen-binding fragment through a linker.

[0024] The linker may be a conjugated bond. The linker may be cleavable or noncleavable. In one embodiment, the linker is a chemical linker. In one embodiment, the linker comprises a covalent bond such an ester bond, an ether bond, an amine bond, an amide bond, a disulphide bond, an imide bond, a sulfone bond, a phosphate bond, a phosphorus ester bond, a peptide bond, a hydrazone bond or a combination thereof. In one embodiment, the linker comprises a hydrophobic poly(ethylene glycol) linker. In one embodiment, the linker comprises a peptide bond.

[0025] In one embodiment, an immunoconjugate is provided that comprises a drug unit and an isolated mAb or antigen-binding fragment that binds specifically to human CD3. In one embodiment, the drug unit in the immunoconjugate may be a chemotherapeutic agent, a growth inhibitory agent, a toxin, or a radioactive isotope. In one embodiment, the drug unit may be a drug unit from class of calicheamicin, an antimitotic agent, a toxin, or a radioactive isotope. In one embodiment, the drug unit comprises a drug unit from class of calicheamicins. Examples of calicheamicins include ozogamicin. In one embodiment, the drug unit comprises an antimitotic agent. Example antimiotic agent includes monomethyl auristatin E. In one embodiment, the drug unit comprises emtansine (DM1).

[0026] In one embodiment, the drug unit is selected from a cytotoxic agent, an immune regulatory reagent, an imaging agent or a combination thereof. In one embodiment, the cytotoxic agent is selected from a growth inhibitory agent or a chemotherapeutic agent from a class of tubulin binders, DNA intercalators, DNA alkylators, enzyme inhibitors, immune modulators, antimetabolite agents, radioactive isotopes, or a combination thereof. In one embodiment, the cytotoxic agent is selected from a calicheamicin, ozogamicin, monomethyl auristatin E, emtansine, a derivative or a combination thereof. In one embodiment, the immune regulatory reagents activate or suppress immune cells, T cell, NK cell, B cell, macrophage, or dendritic cell.

[0027] In one embodiment, the imaging agent may be radionuclide, a florescent agent, a quantum dots, or a combination thereof.

[0028] The application further provides pharmaceutical compositions. In one embodiment, the pharmaceutical composition may include an isolated mAb or antigen-binding fragment disclosed herein and a pharmaceutically acceptable carrier. In one embodiment, the pharmaceutical composition may include an immunoconjugate disclosed herein and a pharmaceutically acceptable carrier.

[0029] In one embodiment, the pharmaceutical composition further comprises radioisotope, radionuclide, a toxin, a therapeutic agent, a chemotherapeutic agent, a drug unit from class of calicheamicin, an antimitotic agent, a radioactive isotope, a therapeutic agent, or a combination thereof. In one embodiment, the therapeutic agent may include an antibody, an enzyme, a chemotherapeutic agent, a growth inhibitory agent, or a combination thereof.

[0030] The application further provides methods for treating a subject with a cancer. In one embodiment, the method includes the steps of administering to the subject an effective amount of the isolated mAb or antigen-binding fragment disclosed herein. In one embodiment, the isolated mAb or antigen-binding fragment binds specifically to human CD3.

[0031] In one embodiment, the method of treating a subject with a cancer may include co-administering a therapeutic agent together with an effective amount of the isolated mAb or antigen-binding fragment having binding specificity to human CD3. In some embodiments, the therapeutic agent may be an antibody, a chemotherapy agent, an enzyme, or a combination thereof. In some embodiments, the therapeutic agent may be capecitabine, cisplatin, trastuzumab, fulvestrant, tamoxifen, letrozole, exemestane, anastrozole, aminoglutethimide, testolactone, vorozole, formestane, fadrozole, letrozole, erlotinib, lafatinib, dasatinib, gefitinib, imatinib, pazopinib, lapatinib, sunitinib, nilotinib, sorafenib, nab-palitaxel, ozogamicin, monomethyl auristatin E, emtansine (DM1), a derivative or a combination thereof.

[0032] Varieties of cancers may be treated using disclosed compositions, isolated mAbs or antigen-binding fragments. In one embodiment, the cancer may include cells that express CD3. Example cancers that may be treated including, without limitation, breast cancer, colorectal cancer, pancreatic cancer, head and neck cancer, melanoma, ovarian cancer, prostate cancer, non-small lung cell cancer, glioma, esophageal cancer, nasopharyngeal cancer, anal cancer, rectal cancer, gastric cancer, bladder cancer, cervical cancer, or brain cancer.

[0033] The subject receiving treatment may be a human. The application further provides a solution that includes an effective concentration of the isolated mAb or an antigen-binding fragment that binds specifically to human CD3. In one embodiment, the solution is blood plasma in a subject.

BRIEF DESCRIPTION OF THE DRAWINGS

[0034] The foregoing and other features of this disclosure will become more fully apparent from the following description and appended claims, taken in conjunction with the accompanying drawings. Understanding that these drawings depict only several embodiments arranged in accordance with the disclosure and are, therefore, not to be considered limiting of its scope, the disclosure will be described with additional specificity and detail through use of the accompanying drawings, in which:

[0035] FIGURE 1 shows the immunization strategy of NZW rabbits with human or cynomolgus CD3;

[0036] FIGURE 2 shows the analysis of rabbit serum for example human and cynomolgus CD3 gamma/epsilon or delta-epsilon Fc-specific IgG before and after immunization;

[0037] FIGURE 3 provides a summary of screening BCC supernatants for rabbit IgG binding human and cynomolgus T cells by FACS and activation of human CD3+ T cells;

[0038] FIGURE 4 shows the binding of example chimeric rabbit/human IgG antibodies to Cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins and human T cell line Jurkat;

[0039] FIGURE 5 shows the FACS analysis of example chimeric rabbit/human CD3-specific antibodies binding to the human T cell line Jurkat;

[0040] FIGURE 6 shows the FACS analysis of example chimeric rabbit/human CD3-specific antibodies binding to the human T cell line Jurkat;

[0041] FIGURE 7 shows the activation of human CD3+ T cells by example chimeric rabbit/human CD3-specific antibodies; and

[0042] FIGURE 8 shows the binding and off-rate analysis of example chimeric rabbit/human or humanized CD3-specific antibodies.

DETAILED DESCRIPTION

[0043] In the following detailed description, reference is made to the accompanying drawings, which form a part hereof. In the drawings, similar symbols typically identify similar components, unless context dictates otherwise. The illustrative embodiments described in the detailed description, drawings, and claims are not meant to be limiting. Other embodiments may be utilized, and other changes may be made, without departing from the spirit or scope of the subject matter presented herein. It will be readily understood that the aspects of the present disclosure, as generally described herein, and illustrated in the Figures, can be arranged, substituted, combined, separated, and designed in a wide variety of different configurations, all of which are explicitly contemplated herein.

[0044] The application provides, among others, isolated antibodies, methods of making such antibodies, bispecific or multi-specific molecules, antibody-drug conjugates and/or immunoconjugates composed from such antibodies or antigen-binding fragments and pharmaceutical compositions containing the antibodies, bispecific or multi-specific molecules, antibody-drug conjugates and/or immunoconjugates, and methods of using such antibodies, conjugates, or compositions for treating a cancer.

[0045] In one aspect, application provides isolated monoclonal antibodies or a fragment thereof that bind to human CD3. The antibodies or a fragment thereof may exhibit one or more desirable functional properties, including without limitation high affinity binding to CD3, the ability to bind to human T cell line Jurkat, the ability to enhance human CD3+T cell activation including proliferation, IFN-γ and/or IL-2 secretion, the ability to stimulate antibody responses and/or the ability to reverse the suppressive function of immunosuppressive cells, such as T regulatory cells. The antibodies or a fragment thereof may be derived from specific heavy and light chain amino acid sequences and/or structural features such as complementarity determining regions (CDRs) composed of specific amino acid sequences as disclosed herein.

[0046] In some embodiments, the antibodies were created by the immunization of rabbits with Human CD3 Delta-Epsilon Fc ("Knobs-into-hole" - KnH) and Gamma-Epsilon Fc (KnH) fusion proteins or HEK 293 cells transiently transfected with human or cynomolgus "CD3 complex" (alpha and beta T cell receptor, gamma, delta, and epsilon accessory chains). Rabbits are known to create antibodies of high affinity,

diversity and specificity (Weber et al. Exp. Mol. Med. 49:e305, which is incorporated herein by reference in its entirety). B cells from immunized animals were cultured *in vitro* and screened for the production of anti-CD3 antibodies. The antibody variable genes were isolated using recombinant DNA techniques and the resulting antibodies were expressed recombinantly and further screened for desired features such as the ability to bind to human and Cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins, to bind to human T cell line Jurkat and Cynomolgus T cell line HSC-F, and the ability to active purified human CD3+ T cells. This general method of antibody discovery is similar to that described in Seeber et al. PLOS One. 9:e86184 (2014), which is incorporated herein by reference in its entirety.

[0047] The term "antibody" is used in the broadest sense and specifically covers single monoclonal antibodies (including agonist and antagonist antibodies), antibody compositions with polyepitopic specificity, as well as antibody fragments (e.g., Fab, $F(ab')_2$, and Fv), so long as they exhibit the desired biological activity. In some embodiments, the antibody may be monoclonal, polyclonal, chimeric, single chain, bispecific or bi-effective, and humanized antibodies, as well as active fragments thereof. Examples of active fragments of molecules that bind to known antigens include Fab, $F(ab')_2$, scFv and Fv fragments, as well as the products of a Fab immunoglobulin expression library and epitope-binding fragments of any of the antibodies and fragments mentioned above. In some embodiments, antibody may include immunoglobulin molecules and immunologically active portions of immunoglobulin molecules, i.e. molecules that contain a binding site with an immunologic binding specificity to an antigen. The immunoglobulin can be of any type (IgG, IgM, IgD, IgE, IgA and IgY) or class (IgG1, IgG2, IgG3, IgG4, IgA1 and IgA2) or subclasses of immunoglobulin molecule. In one embodiment, the antibody may be a whole antibody and any antigen-binding fragment derived from the whole antibody. A typical antibody refers to heterotetrameric protein comprising typically of two heavy (H) chains and two light (L) chains. Each heavy chain is comprised of a heavy chain variable domain (abbreviated as VH) and a heavy chain constant domain. Each light chain is comprised of a light chain variable domain (abbreviated as VL) and a light chain constant domain. The VH and VL regions can be further subdivided into domains of hypervariable complementarity determining regions (CDR), and more conserved regions called framework regions (FR). Each variable domain (either VH or VL) is typically composed of three CDRs and four FRs, arranged in the following order: FR1, CDR1, FR2, CDR2, FR3, CDR3, FR4 from amino-terminus to carboxy-terminus. Within the variable regions of the light and heavy chains there are binding regions that interacts with the antigen.

[0048] 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 (epitope) on the antigen. In addition to their specificity, the monoclonal antibodies are advantageous in that they are synthesized by the hybridoma culture, uncontaminated by other immunoglobulins. 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 disclosure may be made by the hybridoma method first described by Kohler & Milstein, *Nature,* 256:495 (1975), which is incorporated herein by reference in its entirety, or may be made by recombinant DNA methods (see, e.g., U.S. Pat. No. 4,816,567, which is incorporated herein by reference in its entirety).

[0049] Monoclonal antibodies can be produced using various methods including mouse hybridoma or phage display (see Siegel. Transfus. Clin. Biol. 9:15-22 (2002), which is incorporated herein by reference in its entirety) or from molecular cloning of antibodies directly from primary B cells (see Tiller. New Biotechnol. 28:453-7 (2011), which is incorporated herein by reference in its entirety).

[0050] In some embodiments, the monoclonal antibodies may 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 or belonging to a particular antibody class or subclass, while the remainder of the chain(s) is identical with or homologous to corresponding sequences in antibodies derived from another species or belonging to another antibody class or subclass, as well as fragments of such antibodies, so long as they exhibit the desired biological activity (U.S. Pat. No. 4,816,567 (Cabilly et al.); and Morrison et al., *Proc. Natl. Acad. Sci. USA,* 81:6851-6855 [1984]).

[0051] The term "antigen- or epitope-binding portion or fragment" refers to fragments of an antibody that are capable of binding to an antigen (for example, CD3). These fragments may be capable of the antigen-binding function and additional functions of the intact antibody. Examples of binding fragments include, but are not limited to, a single-chain Fv fragment (scFv) consisting of the VL and VH domains of a single arm of an antibody connected in a single polypeptide chain by a synthetic linker, or a Fab fragment that is a monovalent fragment consisting of the VL, constant light (CL), VH and constant heavy 1 (CH1) domains. Antibody fragments can be even smaller sub-fragments and can consist of domains as small as a single CDR domain. In one embodiment, the single CDR domain may be the CDR3 regions from either the VL and/or VH domains (for example, see Beiboer et al., J. Mol. Biol. 296:833-49 (2000), which is incorporated herein by reference in its entirety). Antibody fragments are produced using conventional methods known to those skilled in the art. The antibody fragments are can be screened for utility using the same techniques employed with whole antibodies.

[0052] The "antigen- or epitope-binding portion or fragment" can be derived from an antibody of the present disclosure by a number of art-known techniques. For example, purified monoclonal antibodies can be cleaved with an enzyme, such as pepsin, and subjected to HPLC gel filtration. The appropriate fraction containing Fab fragments can then be collected and concentrated by membrane filtration and the like. For further description of general techniques for the isolation of active fragments of antibodies, see for example, Khaw, B. A. et al. J. Nucl. Med. 23:1011-1019 (1982); Rousseaux et al. Methods Enzymology, 121:663-69, Academic Press, 1986, which are incorporated herein by reference in their entireties.

[0053] Pepsin digestion of antibodies produces two identical antigen-binding fragments, called "Fab" fragments, each with a single antigen-binding site, and a residual "Fc" fragment, whose name reflects its ability to crystallize readily. Pepsin treatment yields an $F(ab')_2$ fragment that has two antigen combining sites and is still capable of cross-linking antigen.

[0054] The Fab fragment may also contain the constant domain of the light chain and the first constant domain (CH1) of the heavy chain. Fab' fragments differ from Fab fragments by the addition of a few residues at the carboxy terminus of the heavy chain CH1 domain including one or more cysteines from the antibody hinge region. Fab'-SH is the designation herein for Fab' in which the cysteine residue(s) of the constant domains bear a free thiol group. $F(ab')_2$ antibody fragments originally were produced as pairs of Fab' fragments that have hinge cysteines between them. Other chemical couplings of antibody fragments known in the field are also useful in this application.

[0055] "Fv" is the minimum antibody fragment that contains a complete antigen recognition and binding site. This region consists of a dimer of one heavy and one light chain variable domain in tight, non-covalent association. It is in this configuration that the three CDRs of each variable domain interact to define an antigen-binding site on the surface of the VH-VL dimer. Collectively, the six CDRs confer antigen-binding specificity to the antibody. However, even a single variable domain (or half of a Fv comprising only three CDRs specific for an antigen) has the ability to recognize and bind the antigen, although at a lower affinity than the entire binding site.

[0056] The "light chains" of antibodies (immunoglobulins) from any vertebrate species can be assigned to one of two clearly distinct types, called kappa and lambda (λ), based on the amino acid sequences of their constant domains.

[0057] Depending on the amino acid sequence of the constant domain of their heavy chains, immunoglobulins can be assigned to different classes. There are five major classes of immunoglobulins: IgA, IgD, IgE, IgG and IgM, and several of these may be further divided into subclasses (isotypes), e.g., IgG-1, IgG-2, IgG-3, and IgG-4; IgA-1 and IgA-2. The heavy chain constant domains that correspond to the different classes of immunoglobulins are called α, delta, epsilon, γ, and μ, respectively. The subunit structures and three-dimensional configurations of different classes of immunoglobulins are well known.

[0058] A "humanized antibody" refers to a type of engineered antibody having its CDRs derived from a non-human donor immunoglobulin, the remaining immunoglobulin-derived parts of the molecule being derived from one (or more) human immunoglobulin(s). In addition, framework support residues may be altered to preserve binding affinity. Methods to obtain "humanized antibodies" are well known to those skilled in the art. (see, e.g., Queen et al., Proc. Natl Acad Sci USA, 86:10029-10032 (1989), Hodgson et al., Bio/Technology, 9:421 (1991)). In one embodiment, the "humanized antibody" may be obtained by a genetic engineering approach that enables production of affinity-matured humanlike polyclonal antibodies in large animals such as, for example, rabbits (see, e.g. U.S. Pat. No. 7,129,084).

[0059] The terms "polypeptide", "peptide", and "protein", as used herein, are interchangeable and are defined to mean a biomolecule composed of amino acids linked by a peptide bond.

[0060] The terms “a”, “an” and “the” as used herein are defined to mean “one or more” and include the plural unless the context is inappropriate.

[0061] By “isolated” is meant a biological molecule free from at least some of the components with which it naturally occurs. "Isolated," when used to describe the various polypeptides disclosed herein, means a polypeptide that has been identified and separated and/or recovered from a cell or cell culture from which it was expressed. Ordinarily, an isolated polypeptide will be prepared by at least one purification step. An "isolated antibody," refers to an antibody which is substantially free of other antibodies having different antigenic specificities.

[0062] "Recombinant" means the antibodies are generated using recombinant nucleic acid techniques in exogeneous host cells.

[0063] The term “antigen” refers to an entity or fragment thereof which can induce an immune response in an organism, particularly an animal, more particularly a mammal including a human. The term includes immunogens and regions thereof responsible for antigenicity or antigenic determinants.

[0064] Also as used herein, the term “immunogenic” refers to substances which elicit or enhance the production of antibodies, T-cells or other reactive immune cells directed against an immunogenic agent and contribute to an immune response in humans or animals. An immune response occurs when an individual produces sufficient antibodies, T-cells and other reactive immune cells against administered immunogenic compositions of the present disclosure to moderate or alleviate the disorder to be treated.

[0065] "Specific binding" or "specifically binds to" or is "specific for" a particular antigen or an epitope means binding that is measurably different from a non-specific interaction. Specific binding can be measured, for example, by determining binding of a molecule compared to binding of a control molecule, which generally is a molecule of similar structure that does not have binding activity. For example, specific binding can be determined by competition with a control molecule that is similar to the target.

[0066] Specific binding for a particular antigen or an epitope can be exhibited, for example, by an antibody having a KD for an antigen or epitope of at least about 10^{-4} M, at least about 10^{-5} M, at least about 10^{-6} M, at least about 10^{-7} M, at least about 10^{-8} M, at least about 10^{-9}, alternatively at least about 10^{-10} M, at least about 10^{-11} M, at least about 10^{-12} M, or greater, where KD refers to a dissociation rate of a particular antibody-antigen interaction. Typically, an antibody that specifically binds an antigen will have a KD that is 20-, 50-, 100-, 500-, 1000-, 5,000-, 10,000- or more times greater for a control molecule relative to the antigen or epitope.

[0067] Also, specific binding for a particular antigen or an epitope can be exhibited, for example, by an antibody having a KA or Ka for an antigen or epitope of at least 20-, 50-, 100-, 500-, 1000-, 5,000-, 10,000- or more times greater for the epitope relative to a control, where KA or Ka refers to an association rate of a particular antibody-antigen interaction.

[0068] "Homology" between two sequences is determined by sequence identity. If two sequences which are to be compared with each other differ in length, sequence identity preferably relates to the percentage of the nucleotide residues of the shorter sequence which are identical with the nucleotide residues of the longer sequence. Sequence identity can be determined conventionally with the use of computer programs. The deviations appearing in the comparison between a given sequence and the above-described sequences of the disclosure may be caused for instance by addition, deletion, substitution, insertion or recombination.

[0069] The application further provides immune-conjugates containing a drug unit linked to the mAbs or their fragments thereof as disclosed herein through a linker. The linker may be cleavable or noncleavable. In one embodiment, the linker is a chemical linker. In one embodiment, the linker comprises a covalent bond such as an ester bond, an ether bond, an amid bond, a disulphide bond, an imide bond, a sulfone bond, a phosphate bond, a phosphorus ester bond, a peptide bond, or a combination thereof. In one embodiment, the linker comprises a hydrophobic poly(ethylene glycol) linker. In one embodiment, the linker comprises a peptide bond.

[0070] The drug unit may include a chemotherapeutic agent, a growth inhibitory agent, a drug unit from class of calicheamicin, an antimitotic agent, a toxin, a radioactive isotope, a therapeutic agent, or a combination thereof. In one embodiment, the therapeutic agent comprises an antibody, a chemotherapy agent, an enzyme, or a combination thereof.

[0071] The application further provides pharmaceutical compositions. In one embodiment, the pharmaceutical composition comprises the mAbs or their fragments thereof, as disclosed herein, and a pharmaceutically acceptable carrier. In one embodiment, pharmaceutical composition comprises the immunoconjugate, as disclosed herein, and a pharmaceutically acceptable carrier.

[0072] The antibodies, their fragment thereof or the immune-conjugate can be prepared in a physiologically acceptable formulation and may comprise a pharmaceutically acceptable carrier, diluent and/or excipient using known techniques. For example, the antibody according to the disclosure may include any functionally equivalent antibody or functional parts thereof, in particular, the monoclonal antibody including any functionally equivalent antibody or functional parts thereof is combined with a pharmaceutically acceptable carrier, diluent and/or excipient to form a therapeutic composition. Suitable pharmaceutical carriers, diluents and/or excipients are well known in the art and include, for example, phosphate buffered saline solutions, water, emulsions such as oil/water emulsions, various types of wetting agents, sterile solutions, etc. In some embodiments, the formulation of the pharmaceutical composition can be accomplished according to standard methodology know to those of ordinary skill in the art.

[0073] Further biologically active agents may be present in the pharmaceutical composition of the disclosure dependent on its the intended use. In one embodiment, the biologically active agent may include capecitabine, cisplatin, trastuzumab, fulvestrant, tamoxifen, letrozole, exemestane, anastrozole, aminoglutethimide, testolactone, vorozole, formestane, fadrozole, letrozole, erlotinib, lafatinib, dasatinib, gefitinib, imatinib, pazopinib, lapatinib, sunitinib, nilotinib, sorafenib, nab-palitaxel,

calicheamicin, antimitotic agent, monomethyl auristatin E, emtansine, ozogamicin, or a derivative or a combination thereof.

[0074] In another aspect, the application provides methods for treating a subject using anti-CD3 antibodies or other molecules containing the antigen-binding portion of an anti-CD3 antibody for activation of human CD3+ T cells. In some embodiments, the methods include the steps of using the antibodies to stimulate a protective autoimmune response, to modify an immune response or to stimulate antigen-specific immune responses. In some embodiments, the methods include administering the disclosed composition into a subject for treating a cancer.

[0075] In some embodiments, the methods include the step of administering an effective amount of pharmaceutical composition disclosed thereof to a subject in need of such treatment. The compositions may be administered to a subject in the form of a solid, liquid or aerosol at a suitable, pharmaceutically effective dose. Examples of solid compositions include pills, creams, and implantable dosage units. Pills may be administered orally. Therapeutic creams may be administered topically. Implantable dosage units may be administered locally, for example, at a tumor site, or may be implanted for systematic release of the therapeutic composition, for example, subcutaneously. Examples of liquid compositions include formulations adapted for injection intramuscularly, subcutaneously, intravenously, intra-arterially, and formulations for topical and intraocular administration. Examples of aerosol formulations include inhaler formulations for administration to the lungs.

[0076] The compositions may be administered by standard routes of administration. In general, the composition may be administered by topical, oral, rectal, nasal, interdermal, intraperitoneal, or parenteral (for example, intravenous, subcutaneous, or intramuscular) routes.

[0077] In one embodiment, the administration can be parenterally, e.g. intravenously. Preparations for parenteral administration include sterile aqueous or non-aqueous solutions, suspensions and emulsions. Non-aqueous solvents include without being limited to it, propylene glycol, polyethylene glycol, vegetable oil such as olive oil, and injectable organic esters such as ethyl oleate. Aqueous solvents may be chosen from the group consisting of water, alcohol/aqueous solutions, emulsions or suspensions including saline and buffered media. Parenteral vehicles include sodium chloride solution, Ringer's dextrose, dextrose and sodium chloride, lactated Ringer's, or fixed oils. Intravenous vehicles include fluid and nutrient replenishers, electrolyte replenishers (such as those based on Ringer's dextrose) and others. Preservatives may also be present such as, for example, antimicrobials, anti-oxidants, chelating agents, inert gases, etc.

[0078] In some embodiments, the composition may be incorporated into sustained release matrices such as biodegradable polymers, the polymers being implanted in the vicinity of where delivery is desired, for example, at the site of a tumor. In one embodiment, a sustained release matrix may be used. The matrix may be made of materials, usually polymers which are degradable by enzymatic or acid/base hydrolysis or by dissolution. Once inserted into the body, the matrix is acted upon by enzymes and body fluids. The sustained release matrix desirably is chosen by biocompatible materials such as liposomes, polylactides (polylactide acid), polyglycolide (polymer of glycolic acid), polylactide co-

glycolide (copolymers of lactic acid and glycolic acid), polyanhydrides, poly(ortho)esters, polypeptides, hyaluronic acid, collagen, chondroitin sulfate, carboxylic acids, fatty acids, phospholipids, polysaccharides, nucleic acids, polyamino acids, amino acids such phenylalanine, tyrosine, isoleucine, polynucleotides, polyvinyl propylene, polyvinylpyrrolidone and silicone. A preferred biodegradable matrix is a matrix of one of either polylactide, polyglycolide, or polylactide co-glycolide (co-polymers of lactic acid and glycolic acid).

[0079] The pharmaceutical composition may further comprise proteinaceous carriers such as, for example, serum albumin or immunoglobulin, particularly of human origin. In some embodiments, proteinaceous pharmaceutically active matter may be present in amounts between 1 ng and 10 mg per dose. In some embodiment, the regime of administration may be in the range of between 0.1 μg and 10 mg of the antibody according to the disclosure, particularly in a range 1.0 μg to 1.0 mg, and more particularly in a range of between 1.0 μg and 100 μg, with all individual numbers falling within these ranges also being part of the disclosure. If the administration occurs through continuous infusion a more proper dosage may be in the range of between 0.01 μg and 10 mg units per kilogram of body weight per hour with all individual numbers falling within these ranges also being part of the disclosure.

[0080] The method may include administration of a single dose, administration of repeated doses at predetermined time intervals, and sustained administration for a predetermined period of time. It is well known to those of ordinary skill in the art that the dosage of the composition will depend on various factors such as, for example, the condition of being treated, the particular composition used, and other clinical factors such as weight, size, sex and general health condition of the patient, body surface area, the particular compound or composition to be administered, other drugs being administered concurrently, and the route of administration.

[0081] The term "therapeutically effective amount" refers to the amount of antibody which, when administered to a human or animal, elicits a response which is sufficient to result in a therapeutic effect in said human or animal. The effective amount is readily determined by one of ordinary skill in the art following routine procedures.

[0082] The composition may be administered in combination with other compositions comprising an biologically active substance or compound. In one embodiment, the biologically active substance or compound may include compounds against oxidative stress, anti-apoptotic compounds, metal chelators, inhibitors of DNA repair such as pirenzepin and metabolites, 3-amino-1-propanesulfonic acid (3APS), 1,3-propanedisulfonate (1,3PDS), secretase activators, β- and γ-secretase inhibitors, tau proteins, neurotransmitter, β-sheet breakers, anti-inflammatory molecules, "atypical antipsychotics" such as, for example clozapine, ziprasidone, risperidone, aripiprazole or olanzapine or cholinesterase inhibitors (ChEIs) such as tacrine, rivastigmine, donepezil, and/or galantamine and other drugs and nutritive supplements such as, for example, vitamin B12, cysteine, a precursor of acetylcholine, lecithin, choline, Ginkgo biloba, acyetyl-L-carnitine, idebenone, propentofylline, or a xanthine derivative.

[0083] The present disclosure may be understood more readily by reference to the following detailed description of specific embodiments included herein. Although the present disclosure has been

described with reference to specific details of certain embodiments thereof, it is not intended that such details should be regarded as limitations upon the scope of the disclosure.

[0084] EXAMPLES

[0085] Example 1: Generation of Anti-CD3 Antibodies

[0086] Monoclonal antibodies against human CD3 were developed by immunizing New Zealand white rabbits. As shown in FIGURE 1, animals were immunized with recombinant Human CD3 Delta-Epsilon Fc ("Knobs-into-hole" - KnH) and Gamma-Epsilon Fc (KnH) fusion proteins or HEK 293 cells transiently transfected with human or cynomolgus "CD3 complex" (alpha and beta T cell receptor, gamma, delta, and epsilon accessory chains) mixed 1:1 v/v with Complete or incomplete Freund's adjuvant (Cohort 1) or Titermax Gold (Cohort 2) alternating with Alhydrogel 2% (Alum) plus CpG 2007 and were performed by subcutaneous injection. Subsequent boosts were performed every 7 days through day 51 (Cohort 1) or day 68 (Cohort 2).

[0087] On week 5 the serum from the animals was tested for CD3 Delta-Epsilon Fc ("Knobs-into-hole" - KnH) and Gamma-Epsilon Fc (KnH) fusion protein titer by ELISA. Serum from each rabbit was obtained before immunization as a negative control. After immunization serum was again collected from each animal and compared to the pre-immunization serum from the same animal for the presence of CD3 Delta, Gamma, or Epsilon-specific IgG antibodies. As shown in FIGURE 2 all animals immunized with human and cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins developed detectable titers of IgG specific for human CD3 Delta-Epsilon Fc (plate 1), human CD3 Gamma-Epsilon Fc (plate 2), Cynomolgus CD3 Delta-Epsilon Fc (plate 3), Cynomolgus CD3 Gamma-Epsilon Fc CD3-Fc fusion proteins.

[0088] One rabbit in cohort No.2, 6235R, which was immunized with human or cynomolgus CD3 transfected cells, showed serum IgG binding to the human and cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins, indicating that the specificity was for the Delta, Gamma, or Epsilon part of the Fc fusion proteins. Antigen-specific B cells were enriched using biotinylated cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins and seeded at limit dilution into multiple 96-well tissue culture plates and cultured for 9 days to allow their differentiation into plasma cells and for secretion of antibodies. The supernatants from these plasma cell cultures were screened by ELISA, flow cytometry (FACS), and functional assay for the presence of CD3-specific antibodies in a series of assays as listed below:

1. Binding to human CD3 Delta, Gamma, or Epsilon Fc fusion proteins – detection of Delta, Gamma, or Epsilon-specific IgG ELISA

2. Binding to Cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins – detection of Delta, Gamma, or Epsilon-specific IgG ELISA

3. Binding to human IgG – counter-screen for detection of human IgG Fc-specific IgG ELISA

4. Binding to human T cell line Jurkat – detection of IgG which is specific for CD3 Delta, Gamma, or Epsilon FACS

5. Binding to Cynomolgus T cell line HSC-F – detection of IgG which is specific for CD3 Delta, Gamma, or Epsilon FACS

6. Binding to non-transfected human embryonic kidney (HEK) 293 cells – counter-screen for detection of IgG specific for unknown target(s) on cells that do not express human or cynomolgus CD3 Delta, Gamma, or Epsilon

7. Activation of purified human CD3+ T cells – detection of IL-2 secretion ELISA

[0089] On day 9 of B cell culture the supernatants were separated from the B cells and stored in a separate plate for later analysis. RNAlater tissue storage reagent was added to each well in the B cell culture plate to preserve the RNA in the B cells for RT-PCR amplification of antibody variable regions.

[0090] After the BCC supernatants were screened for binding as in points 1-3 above 23 wells were identified that contained IgG that specifically recognized human and cynomolgus CD3 epsilon. This set of 23 wells were then screened for binding human and cynomolgus T cells, as well as activation of human T cells, and the results are shown in FIGURE 3.

[0091] This set of 23 BCC wells identified through ELISA and FACS screening as having the desired antibodies we advanced to molecular "rescue" of the antibody variable regions. The light and heavy chain variable sequences were amplified by multiplex RT-PCR using degenerate primers designed to anneal to leader sequences and the constant regions of rabbit IgG and rabbit kappa sequences. Secondary PCR was performed separately for the light and heavy chains using nested primers containing restriction sites. Amplicons from the variable heavy chain PCR were cloned into an expression vector containing human IgG1. Light chain amplicons were cloned into an expression vector containing human IgK. Resulting clones were sequenced and analyzed.

[0092] The heavy and light chain expression plasmids generated from each well were transiently co-transfected to produce rabbit/human chimeric antibodies. Recombinant antibody supernatants were confirmed to contain antibodies specific for Cynomolgus CD3 Delta, Gamma, or Epsilon using bio-layer interferometry analysis on a ForteBio Octet Red 96 instrument. Anti-human Fc biosensors (Pall ForteBio) were used to capture antibodies in the supernatants. Association to Cynomolgus CD3 Delta, Gamma, or Epsilon was observed by real-time interferometry by placing the biosensors in wells containing recombinant human ROR1 extracellular domain protein. Dissociation was measured after transfer of the biosensors into wells containing 10X kinetics buffer (Pall ForteBio). The software provided by the manufacturer was used to analyze the interferometry data.

[0093] A summary of the primary BCC screening data and the corresponding screening data for 22 recombinant chimeric rabbit/human IgG antibodies is shown in FIGURE 4. From the original 23 BCC wells 22 chimeric antibodies were rescued, 6 showed binding to the Cynomolgus CD3 Delta/Epsilon Fc fusion protein, 5 of 6 showed binding to Cynomolgus CD3 Gamma/Epsilon Fc fusion protein, and 4 of 5 showed binding to the human T cell line Jurkat (FIGURE 5 and FIGURE 4).

[0094] A second round of B cell culture was setup with Cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion protein binding IgG+ B cells from rabbit 6235R sorted at 1 per well into multiple 96-well tissue culture plates and cultured for 9 days to allow their differentiation into plasma cells and for secretion of antibodies. The supernatants from these plasma cell cultures were screened by ELISA, flow cytometry (FACS), and functional assay for the presence of CD3-specific antibodies as shown in the list above. As shown in FIGURE 6, 5 chimeric rabbit/human antibodies were tested for binding to the human T cell line Jurkat and 3 of 5 showed binding.

[0095] From the first BCC 4 chimeric rabbit/human antibodies, and from the second BCC 3 chimeric rabbit/human antibodies, showed binding to the human T cell line Jurkat for a total of 7. These 7 antibodies and 2 additional non-binding antibodies were analyzed for activation of human CD3+ T cells. As shown in FIGURE 7, of the 9 antibodies tested 7 activated human CD3+ T cells, 6 of them strongly, 284A10, 295E9, 299F6, 480A7, 480C8, 485H6.

[0096] The heavy and light chain variable regions for the 6 chimeric rabbit/human IgG antibodies listed above were humanized. Humanized variants for 3 of 6 showed similar binding kinetics to human CD3 Delta/Epsilon Fc and/or Gamma/Epsilon Fc fusion protein by octet analysis which is summarized in FIGURE 8.

SEQUENCE LISTING

SEQ ID NO: 1

284A10 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCTCCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAAGTGCCAGGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAA
GCATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCA
GTGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATT
CTTTCGGCGGAGGCACCGAGGTGGAGTTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGA
TGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAG
TGGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCAC
CTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCAC
CCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 2

284A10 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCTCCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAAGTGCCAGGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAA
GCATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCA
GTGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATT
CTTTCGGCGGAGGCACCGAGGTGGAGTTCAAA

SEQ ID NO: 3

284A10 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPPSVSAAVGGTVTIKCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRTYVNSFGGGTEVEFKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL
QSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 4

284A10 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPPSVSAAVGGTVTIKCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRTYVNSFGGGTEVEFK

SEQ ID NO: 5

284A10 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
TTCACCATCAGTACCAATGCAATGAGCTGGGTCCGCCAGGTTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCATT
ACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAACAACATTTGGGGCCCAGGCACCCTCGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGGT
CTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTTC
CCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACAG
TCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGCA
ACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACAT
GCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCCT
CATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCAA
CTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTACC
GTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAACA
AAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACACCC
TGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGCG
ACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTCC
GACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATGCT
CCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 6

284A10 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
TTCACCATCAGTACCAATGCAATGAGCTGGGTCCGCCAGGTTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCATT
ACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAACAACATTTGGGGCCCAGGCACCCTCGTCACCGTCTCGAGC

SEQ ID NO: 7

284A10 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGFTISTNAMSWVRQVPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSNNIWGPGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAPS
VFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNG
KEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPP
VLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 8

284A10 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGFTISTNAMSWVRQVPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSNNIWGPGTLVTVSS

SEQ ID NO: 9

299F6 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCATCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAAG
CATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCAG
CGGCGTGCAGTGTGACGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTGAATG
CTTTCGGCGGAGGGACCGAGGTGGGGGTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTG
ATGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACA
GTGGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCA
CCTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCA
CCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 10

299F6 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCATCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAAG
CATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCAG
CGGCGTGCAGTGTGACGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTGAATG
CTTTCGGCGGAGGGACCGAGGTGGGGGTCAAA

SEQ ID NO: 11

299F6 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPSSVSAAVGGTVTINCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVFKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL
QSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 12

299F6 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPSSVSAAVGGTVTINCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVFK

SEQ ID NO: 13

299F6 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
TTCACCATCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAACTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAACAACATTTGGGGCCCAGGGACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGG
TCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTT
CCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACA
GTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGC
AACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACA
TGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCC
TCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCA
ACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTA
CCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAA
CAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACAC
CCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGC
GACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTC
CGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATG
CTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 14

299F6 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
TTCACCATCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAACTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAACAACATTTGGGGCCCAGGGACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 15

299F6 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGFTISSNAMSWVRQAPGKGLEWIGVITGRDITYYANWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSNNIWGPGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAPS
VFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNG
KEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPP
VLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 16

299F6 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGFTISSNAMSWVRQAPGKGLEWIGVITGRDITYYANWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSNNIWGPGTLVTVSS

SEQ ID NO: 17

480C8 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCATCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAAG
CATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCAG
CGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATG
CTTTCGGCGGAGGGACCGAGGTGGAGTTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGA
TGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAG
TGGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCAC
CTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCAC
CCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 18

480C8 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCATCCTCCGTGTCTGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAAG
CATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCAG
CGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATG
CTTTCGGCGGAGGGACCGAGGTGGAGTTCAAA

SEQ ID NO: 19

480C8 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPSSVSAAVGGTVTINCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRTYVNAFGGGTEVEFKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL
QSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 20

480C8 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPSSVSAAVGGTVTINCQASESISSWLAWYQQKPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRTYVNAFGGGTEVEFK

SEQ ID NO: 21

480C8 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTAATAGTAAGAACATTTGGGGCCAAGGCACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGG
TCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTT
CCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACA
GTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGC
AACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACA
TGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCC
TCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCA
ACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTA
CCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAA
CAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACAC
CCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGC
GACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTC
CGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATG
CTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 22

480C8 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACGCCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTAATAGTAAGAACATTTGGGGCCAAGGCACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 23

480C8 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAINSKNIWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAPS
VFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNG

KEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPP
VLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 24

480C8 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAINSKNIWGQGTLVTVSS

SEQ ID NO: 25

295E9 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGGAGGCAGCTGTGGGAGGCACAGTCACAATCAATTGCCAAGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAGACCAGGGCAGCCTCCCAAACTCCTGATCTACGAA
GCATCCAAACTGGCGTCTGGGGTCCCATCGCGGTTCAAAGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATC
AGCGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAA
TGCTTTCGGCGGAGGGACCGAGGTGGTGGTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCT
GATGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTAC
AGTGGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGC
ACCTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTC
ACCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 26

295E9 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGGAGGCAGCTGTGGGAGGCACAGTCACAATCAATTGCCAAGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAGACCAGGGCAGCCTCCCAAACTCCTGATCTACGAA
GCATCCAAACTGGCGTCTGGGGTCCCATCGCGGTTCAAAGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATC
AGCGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAA
TGCTTTCGGCGGAGGGACCGAGGTGGTGGTCAAA

SEQ ID NO: 27

295E9 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPASVEAAVGGTVTINCQASESISSWLAWYQQRPGQPPKLLIYEASKLASGVPSRFKGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVVKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL
QSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 28

295E9 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPASVEAAVGGTVTINCQASESISSWLAWYQQRPGQPPKLLIYEASKLASGVPSRFKGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVVK

SEQ ID NO: 29

295E9 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCACTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGTAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATTAATTACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCCAGGCACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGG
TCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTT
CCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACA
GTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGC
AACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACA
TGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCC
TCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCA
ACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTA
CCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAA
CAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACAC
CCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGC
GACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTC
CGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATG
CTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 30

295E9 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCACTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGTAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATTAATTACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCCAGGCACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 31

295E9 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDINYYASWAKGRFTISKTSSTTVDLK
MTSLTAEDSATYFCARDGGSSAITSKNIWGPGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSW
NSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAP
SVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNG
KEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPP
VLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 32

295E9 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDINYYASWAKGRFTISKTSSTTVDLK
MTSLTAEDSATYFCARDGGSSAITSKNIWGPGTLVTVSS

SEQ ID NO: 33

480A7 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGGAGGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAGACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAA
GCATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCA
GCGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAGGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAAT
GCTTTCGGCGGAGGGACCGAGGTGGTGTTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTG
ATGAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACA
GTGGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCA
CCTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCA
CCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 34

480A7 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGGAGGCAGCTGTGGGAGGCACAGTCACCATCAATTGCCAAGCC
AGTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAGACCAGGGCAGCCTCCCAAGCTCCTGATCTACGAA
GCATCCAAACTGGCATCTGGGGTCCCATCGCGGTTCAGTGGCAGTGGATCTGGGACAGAGTTCACTCTCACCATCA
GCGACCTGGAGTGTGCCGATGCTGCCACTTACTACTGTCAGGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAAT
GCTTTCGGCGGAGGGACCGAGGTGGTGTTCAAA

SEQ ID NO: 35

480A7 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPASVEAAVGGTVTINCQASESISSWLAWYQQRPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVFKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL
QSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 36

480A7 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPASVEAAVGGTVTINCQASESISSWLAWYQQRPGQPPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISDLEC
ADAATYYCQGYFYFISRSYVNAFGGGTEVVFK

SEQ ID NO: 37

480A7 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCCGGGCACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGG
TCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTT
CCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACA
GTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGC
AACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACA
TGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCC
TCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCA
ACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTA
CCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAA
CAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACAC
CCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGC
GACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTC
CGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATG
CTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 38

480A7 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG

GTGGATCTGAAAATGACCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCCGGGCACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 39

480A7 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSKNIWGPGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNS
GALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAPSV
FLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKE
YKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVL
DSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 40

480A7 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
TSLTAEDSATYFCARDGGSSAITSKNIWGPGTLVTVSS

SEQ ID NO: 41

485H6 CHIMERIC LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGTCTGAACCTGTGGGAGGCACAGTCACCATCAAGTGCCAGGCC
AGTGAGAGCATTAATAGTAATGTAGCCTGGTATCAGCAGAAACCAGGGCAGCGTCCCAAGCTCCTGATCTATGAT
TCATCCACTCTGGAATCTGGGGTCCCATCGCGGTTCAAAGGCAGTAGATCTGGGACAGAGTTCACTCTCACCATCA
GCGACCTGGAGTGTGCCGATGCCGCCACTTACTACTGTCAATGTAGTAGTTATGGTAGTAGTTATGTTGGTGGTTT
CGGCGGAGGGACCGAGGTGGTGTTCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGATGA
GCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGTGG
AAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCACCTA
CAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCACCCA
TCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 42

485H6 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GATGTCGTGATGACCCAGACTCCAGCCTCCGTGTCTGAACCTGTGGGAGGCACAGTCACCATCAAGTGCCAGGCC
AGTGAGAGCATTAATAGTAATGTAGCCTGGTATCAGCAGAAACCAGGGCAGCGTCCCAAGCTCCTGATCTATGAT
TCATCCACTCTGGAATCTGGGGTCCCATCGCGGTTCAAAGGCAGTAGATCTGGGACAGAGTTCACTCTCACCATCA
GCGACCTGGAGTGTGCCGATGCCGCCACTTACTACTGTCAATGTAGTAGTTATGGTAGTAGTTATGTTGGTGGTTT
CGGCGGAGGGACCGAGGTGGTGTTCAAA

SEQ ID NO: 43

485H6 CHIMERIC LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQTPASVSEPVGGTVTIKCQASESINSNVAWYQQKPGQRPKLLIYDSSTLESGVPSRFKGSRSGTEFTLTISDLECA
DAATYYCQCSSYGSSYVGGFGGGTEVVFKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS
GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 44

485H6 CHIMERIC LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQTPASVSEPVGGTVTIKCQASESINSNVAWYQQKPGQRPKLLIYDSSTLESGVPSRFKGSRSGTEFTLTISDLECA
DAATYYCQCSSYGSSYVGGFGGGTEVVFK

SEQ ID NO: 45

485H6 CHIMERIC HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGCCCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCAGGGGACCCTCGTCACCGTCTCGAGCGCTAGCACCAAGGGCCCATCGG
TCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTT
CCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACA
GTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGC
AACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAACTCACACA
TGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAAGGACACCC
TCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGTCAAGTTCA
ACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACAGCACGTA
CCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGGTCTCCAA
CAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGGTGTACAC
CCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTATCCCAGC
GACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTC
CGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTCTCATG
CTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 46

485H6 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

CAGTCGCTGGAGGAGTCCGGGGGTCGCCTGGTCACGCCTGGGACACCCCTGACACTCACCTGCACAGTCTCTGGA
ATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAGTCAT
TACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCCGATTCACCATCTCCAAAACCTCGTCGACCACG
GTGGATCTGAAAATGCCCAGTCTGACAGCCGAGGACTCGGCCACCTATTTCTGTGCCAGAGACGGTGGTTCTTCTG
CTATTACTAGTAAGAACATTTGGGGCCAGGGGACCCTCGTCACCGTCTCGAGC

SEQ ID NO: 47

485H6 CHIMERIC HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
PSLTAEDSATYFCARDGGSSAITSKNIWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGAPS
VFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNG
KEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPP
VLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 48

485H6 CHIMERIC HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

QSLEESGGRLVTPGTPLTLTCTVSGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISKTSSTTVDLKM
PSLTAEDSATYFCARDGGSSAITSKNIWGQGTLVTVSS

SEQ ID NO: 49

284A10 HUMANIZED LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GACGTCGTGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG
CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATTC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGAT
GAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGT
GGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCACC
TACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCACC
CATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 50

284A10 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GACGTCGTGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCAATTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG

CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATTC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAA

SEQ ID NO: 51

284A10 HUMANIZED LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DVVMTQSPSTLSASVGDRVTINCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRTYVNSFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS
GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 52

284A10 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DVVMTQSPSTLSASVGDRVTINCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRTYVNSFGGGTKVEIK

SEQ ID NO: 53

284A10 HUMANIZED HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGATTCACCATCAGTACCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTACTAGTAACAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGG
CCCATCGGTCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAG
GACTACTTCCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCT
GTCCTACAGTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCT
ACATCTGCAACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAA
CTCACACATGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAA
GGACACCCTCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGT
CAAGTTCAACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACA
GCACGTACCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGG
TCTCCAACAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGG
TGTACACCCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTA
TCCCAGCGACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGC
TGGACTCCGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCT
TCTCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 54

284A10 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGATTCACCATCAGTACCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTACTAGTAACAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 55

284A10 HUMANIZED HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGFTISTNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISRDNSKNTLY
LQMNSLRAEDTAVYYCARDGGSSAITSNNIWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT
VSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAA
GAPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDW
LNGKEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKT
TPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 56

284A10 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGFTISTNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISRDNSKNTLY
LQMNSLRAEDTAVYYCARDGGSSAITSNNIWGQGTLVTVSS

SEQ ID NO: 57

299F6 HUMANIZED LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GACATCCAGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCACTTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG
CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAATGC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGAT
GAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGT
GGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCACC
TACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCACC
CATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 58

299F6 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GACATCCAGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCACTTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG
CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTAGTTATGTAAATGC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAA

SEQ ID NO: 59

299F6 HUMANIZED LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DIQMTQSPSTLSASVGDRVTITCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRSYVNAFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS
GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 60

299F6 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DIQMTQSPSTLSASVGDRVTITCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRSYVNAFGGGTKVEIK

SEQ ID NO: 61

299F6 HUMANIZED HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGATTCACCATCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAACTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTACTAGTAACAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGGG
CCCATCGGTCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAG
GACTACTTCCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGCT
GTCCTACAGTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACCT
ACATCTGCAACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAAA
CTCACACATGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCAA
GGACACCCTCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGGT
CAAGTTCAACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAACA
GCACGTACCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCGG
TCTCCAACAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAGG

TGTACACCCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTA
TCCCAGCGACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGC
TGGACTCCGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCT
TCTCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 62

299F6 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGATTCACCATCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAACTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTACTAGTAACAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 63

299F6 HUMANIZED HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGFTISSNAMSWVRQAPGKGLEWIGVITGRDITYYANWAKGRFTISRDNSKNTL
YLQMNSLRAEDTAVYYCARDGGSSAITSNNIWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT
VSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAA
GAPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDW
LNGKEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKT
TPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 64

299F6 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGFTISSNAMSWVRQAPGKGLEWIGVITGRDITYYANWAKGRFTISRDNSKNTL
YLQMNSLRAEDTAVYYCARDGGSSAITSNNIWGQGTLVTVSS

SEQ ID NO: 65

480C8 HUMANIZED LIGHT CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GACATCCAGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCACTTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG
CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATGC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAACGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGAT
GAGCAGTTGAAATCTGGAACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGT
GGAAGGTGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGGACAGCACC

TACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCACC
CATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCAACAGGGGAGAGTGT

SEQ ID NO: 66

480C8 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN NUCLEOTIDE SEQUENCE

GACATCCAGATGACCCAGTCTCCTTCCACCCTGTCTGCATCTGTAGGAGACAGAGTCACCATCACTTGCCAAGCCA
GTGAGAGCATTAGCAGTTGGTTAGCCTGGTATCAGCAGAAACCAGGGAAAGCCCCTAAGCTCCTGATCTATGAAG
CATCCAAACTGGCATCTGGGGTCCCATCAAGGTTCAGCGGCAGTGGATCTGGGACAGAATTCACTCTCACCATCAG
CAGCCTGCAGCCTGATGATTTTGCAACTTATTACTGCCAAGGCTATTTTTATTTTATTAGTCGTACTTATGTAAATGC
TTTCGGCGGAGGGACCAAGGTGGAGATCAAA

SEQ ID NO: 67

480C8 HUMANIZED LIGHT CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN KAPPA CONSTANT DOMAIN IS UNDERLINED

DIQMTQSPSTLSASVGDRVTITCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRTYVNAFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS
GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 68

480C8 HUMANIZED LIGHT CHAIN VARIABLE LIGHT CHAIN AMINO ACID SEQUENCE. COMPIMENTARITY DETERMINING REGIONS ARE UNDERLINED

DIQMTQSPSTLSASVGDRVTITCQASESISSWLAWYQQKPGKAPKLLIYEASKLASGVPSRFSGSGSGTEFTLTISSLQPD
DFATYYCQGYFYFISRTYVNAFGGGTKVEIK

SEQ ID NO: 69

480C8 HUMANIZED HEAVY CHAIN FULL-LENGTH NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGAATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTAATAGTAAGAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGCGCTAGCACCAAGG
GCCCATCGGTCTTCCCCCTGGCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAA
GGACTACTTCCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCACACCTTCCCGGC
TGTCCTACAGTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAGCTTGGGCACCCAGACC
TACATCTGCAACGTGAATCACAAGCCCAGCAACACCAAGGTGGACAAGAGAGTTGAGCCCAAATCTTGTGACAAA
ACTCACACATGCCCACCGTGCCCAGCACCTGAAGCCGCGGGGGCACCGTCAGTCTTCCTCTTCCCCCCAAAACCCA
AGGACACCCTCATGATCTCCCGGACCCCTGAGGTCACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCTGAGG
TCAAGTTCAACTGGTACGTGGACGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGGGAGGAGCAGTACAAC

AGCACGTACCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAATGGCAAGGAGTACAAGTGCGCG
GTCTCCAACAAAGCCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAGCCAAAGGGCAGCCCCGAGAACCACAG
GTGTACACCCTGCCCCCATCCCGGGATGAGCTGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCT
ATCCCAGCGACATCGCCGTGGAGTGGGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTG
CTGGACTCCGACGGCTCCTTCTTCCTCTATAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTC
TTCTCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTCTCCGGGT

SEQ ID NO: 70

480C8 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN NUCLEOTIDE SEQUENCE

GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGTCCAGCCTGGGGGGTCCCTGAGACTCTCCTGTGCAGCCTC
TGGAATCGACCTCAGTAGCAATGCAATGAGCTGGGTCCGCCAGGCTCCAGGGAAGGGGCTGGAGTGGATCGGAG
TCATTACTGGTCGTGATATCACATACTACGCGAGCTGGGCGAAAGGCAGATTCACCATCTCCAGAGACAATTCCAA
GAACACGCTGTATCTTCAAATGAACAGCCTGAGAGCCGAGGACACGGCTGTGTATTACTGTGCGAGAGACGGTG
GTTCTTCTGCTATTAATAGTAAGAACATTTGGGGCCAGGGAACCCTGGTCACCGTCTCGAGC

SEQ ID NO: 71

480C8 HUMANIZED HEAVY CHAIN FULL-LENGTH AMINO ACID SEQUENCE. HUMAN GAMMA-1 CONSTANT DOMAIN IS UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISRDNSKNTLY
LQMNSLRAEDTAVYYCARDGGSSAINSKNIWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT
VSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAA
GAPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDW
LNGKEYKCAVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKT
TPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG

SEQ ID NO: 72

480C8 HUMANIZED HEAVY CHAIN VARIABLE HEAVY CHAIN AMINO ACID SEQUENCE. COMPLIMENTARITY DETERMINING REGIONS ARE UNDERLINED

EVQLVESGGGLVQPGGSLRLSCAASGIDLSSNAMSWVRQAPGKGLEWIGVITGRDITYYASWAKGRFTISRDNSKNTLY
LQMNSLRAEDTAVYYCARDGGSSAINSKNIWGQGTLVTVSS

CLAIMS

What is claimed is:

1. An isolated monoclonal antibody (mAb) or antigen-binding fragment thereof having a binding specificity to human CD3, wherein the isolated mAb or antigen-binding fragment comprises an amino acid sequence having an identity with a sequence selected from SEQ ID NO:4, SEQ ID NO:8, SEQ ID NO:12, SEQ ID NO:16, SEQ ID NO:20, SEQ ID NO:24, SEQ ID NO:28, SEQ ID NO:32, SEQ ID NO:36, SEQ ID NO:40, SEQ ID NO:44, SEQ ID NO:48, SEQ ID NO:52, SEQ ID NO:56, SEQ ID NO:60, SEQ ID NO:64, SEQ ID NO:68, or SEQ ID NO:72, wherein the identity is not less than at least 98%.

2. The isolated monoclonal antibody (mAb) or antigen-binding fragment according to Claim 1, wherein the identity is not less than 98%.

3. The isolated mAb or antigen-binding fragment according to Claim 1, having a binding affinity to human CD3 with a Kd not greater than 70nM.

4. The isolated mAb or antigen-binding fragment according to Claim 1, wherein the isolated mAb or antigen-binding fragment is capable of high affinity binding to CD3, enhancing T cell activation, stimulating antibody response, or reversing the suppressive function of an immunosuppressive cell.

5. The isolated mAb or antigen-binding fragment according to Claim 4, wherein the enhancing T-cell activation comprises T-cell proliferation, IFN-γ and/or IL-2 secretion, or a combination thereof.

6. The isolated mAb or antigen-binding fragment according to Claim 4, wherein the immunosuppressive cell comprises a T regulatory cell.

7. The isolated mAb or an antigen-binding fragment thereof according to claim 1, wherein the isolated mAb is a humanized antibody, a chimeric antibody, or a recombinant antibody.

8. The isolated mAb or an antigen-binding fragment thereof according to claim 1, wherein the isolated mAb is an IgG.

9. The isolated mAb or an antigen-binding fragment thereof according to claim 1, wherein the antigen-binding fragment is a Fv, a Fab, a F(ab')2, a scFV, a scFV2 fragment, or a combination thereof.

10. The isolated mAb or an antigen-binding fragment thereof according to claim 1, wherein the isolated mAb is a bispecific antibody, tri-specific antibody, or multi-specific antibody.

11. An IgG1 heavy chains for an isolated mAb or an antigen-binding fragment having a binding specificity to human CD3, comprising an amino acid sequence having at least 90% identity with SEQ ID NO:7, SEQ ID NO:15, SEQ ID NO:23, SEQ ID NO:31, SEQ ID NO:39, SEQ ID NO:47, SEQ ID NO:55, SEQ ID NO:63, or SEQ ID NO:71.

12. A kappa light chain for an isolated mAb or an antigen-binding fragment having a binding specificity to human CD3, comprising an amino acid sequence having at least 90% identity with SEQ ID NO:3, SEQ

ID NO:11, SEQ ID NO:19, SEQ ID NO:27, SEQ ID NO:35, SEQ ID NO:43, SEQ ID NO:51, SEQ ID NO:59, or SEQ ID NO:67.

13. A variable light chain for an isolated mAb or an antigen-binding fragment having a binding specificity to human CD3, comprising an amino acid sequence having at least 90% identity with SEQ ID NO:4, SEQ ID NO:12, SEQ ID NO:20, SEQ ID NO:28, SEQ ID NO:36, SEQ ID NO:44, SEQ ID NO:52, SEQ ID NO:60, or SEQ ID NO: 68.

14. A variable heavy chain for an isolated mAb or an antigen-binding fragment having a binding specificity to human CD3, comprising an amino acid sequence having at least 90% identity with SEQ ID NO:8, SEQ ID NO:16, SEQ ID NO:24, SEQ ID NO:32, SEQ ID NO:40, SEQ ID NO:48, SEQ ID NO:56, SEQ ID NO:64, or SEQ ID NO:72.

15. An isolated nucleic acid encoding the isolated mAb or an antigen-binding fragment according to Claim 1, the IgG1 heavy Chain of Claim 11, the kappa light chain of Claim 12, the variable light chain of Claim 13, or the variable heavy chain of Claim 14.

16. An expression vector comprising the isolated nucleic acid of Claim 15.

17. A host cell comprising the nucleic acid of Claim 15, wherein the host cell is a prokaryotic cell or a eukaryotic cell.

18. A method of producing an antibody comprising culturing the host cell of Claim 17 so that the antibody is produced.

19. An immune-conjugate, comprising the isolated mAb or an antigen-binding fragment thereof according to Claim 1 and a drug unit, wherein the drug unit is linked to the isolated mAb or an antigen-binding fragment through a linker, and wherein the linker comprises a covalent bond selected from an ester bond, an ether bond, an amine bond, an amide bond, a disulphide bond, an imide bond, a sulfone bond, a phosphate bond, a phosphorus ester bond, a peptide bond, a hydrazone bond or a combination thereof.

20. The immune-conjugate according to claim 19, wherein the drug unit comprises a cytotoxic agent, an immune regulatory reagent, an imaging agent or a combination thereof.

21. The immune-conjugate according to claim 20, wherein the cytotoxic agent is selected from a growth inhibitory agent or a chemotherapeutic agent from a class of tubulin binders, DNA intercalators, DNA alkylators, enzyme inhibitors, immune modulators, antimetabolite agents, radioactive isotopes, or a combination thereof.

22. The immune-conjugate according to claim 20, wherein the cytotoxic agent is selected from a calicheamicin, ozogamicin, monomethyl auristatin E, emtansine, a derivative or a combination thereof.

23. The immune-conjugate according to claim 20, wherein the immune regulatory reagents activate or suppress immune cells, T cell, NK cell, B cell, macrophage, or dendritic cell.

24. The immune-conjugate according to claim 19, wherein the imaging agent may be radionuclide, a florescent agent, a quantum dots, or a combination thereof.

25. A pharmaceutical composition, comprising the isolated mAb or an antigen-binding fragment thereof according to Claim 1 and a pharmaceutically acceptable carrier.

26. The pharmaceutical composition of Claim 25, further comprising a chemotherapeutic agent, a growth inhibitory agent, a cytotoxic agent from class of calicheamicin, an antimitotic agent, a toxin, a radioactive isotope, a therapeutic agent, or a combination thereof.

27. A pharmaceutical composition, comprising the immune-conjugate of Claim 19 and a pharmaceutically acceptable carrier.

28. A method of treating a subject with a cancer, comprising administering to the subject an effective amount of the isolated mAb or an antigen-binding fragment thereof according to Claim 1.

30. The method of Claim 28, further comprising co-administering an effective amount of a therapeutic agent, wherein the therapeutic agent comprises an antibody, a chemotherapy agent, an enzyme, or a combination thereof.

32. The method of Claim 28, wherein the subject is a human.

33. A solution comprising an effective concentration of the isolated mAb or an antigen-binding fragment thereof according to Claim 1, wherein the solution is blood plasma in a subject.

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 1/8

FIGURE 1 shows the immunization strategy of NZW rabbits with human or cynomolgus CD3 using two cohorts in 1A and 1B.

Day	Cohort	Antigen	Adjuvant
0	1	Human CD3 Delta-Epsilon Fc (KnH) and Gamma-Epsilon Fc (KnH)	Freund's
7	1	Human CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
14	1	Cynomolgous CD3 Delta-Epsilon Fc and (KnH) Gamma-Epsilon Fc (KnH)	Incomplete Fruend's
21	1	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
28	1	Human CD3 Delta-Epsilon Fc (KnH) and Gamma-Epsilon Fc (KnH)	Incomplete Fruend's
35	1	Human CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
42	1	Human CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
51	1	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
55		Harvest	NA

FIGURE 1A

Day	Cohort	Antigen	Adjuvant
0	2	Human CD3 TcR transfected cells	Titermax
7	2	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
14	2	Human CD3 TcR transfected cells	Titermax
21	2	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
28	2	Human CD3 TcR transfected cells	Titermax
37	2	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
47	2	Human CD3 TcR transfected cells	Titermax
54	2	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
61	2	Cynomolgous CD3 TcR transfected cells	Titermax
68	2	Cynomolgous CD3 TcR transfected cells	Alyhrodgel 2% + CpG 2007
72	2	Harvest	NA

FIGURE 1B

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 2/8

FIGURE 2 shows the analysis of rabbit serum analyzed by ELISA human or cynomolgus CD3 gamma-epsilon or delta-epsilon Fc-specific IgG before and after immunization. 2A: Analysis of rabbit serum analyzed by ELISA for human CD3 delta-epsilon Fc specific IgG before and after immunization; 2B: Analysis of rabbit serum analyzed by ELISA for human gamma-epsilon Fc-specific IgG before and after immunization; 2C: Analysis of rabbit serum analyzed by ELISA for cynomolgus CD3 delta-epsilon Fc-specific IgG before and after immunization; and 2D: Analysis of rabbit serum analyzed by ELISA for cynomolgus CD3 gamma-epsilon Fc-specific IgG before and after immunization.

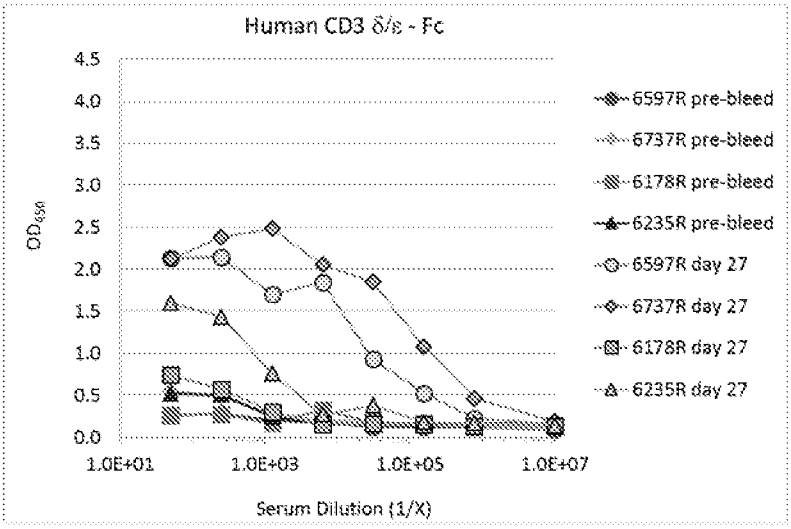


FIGURE 2A

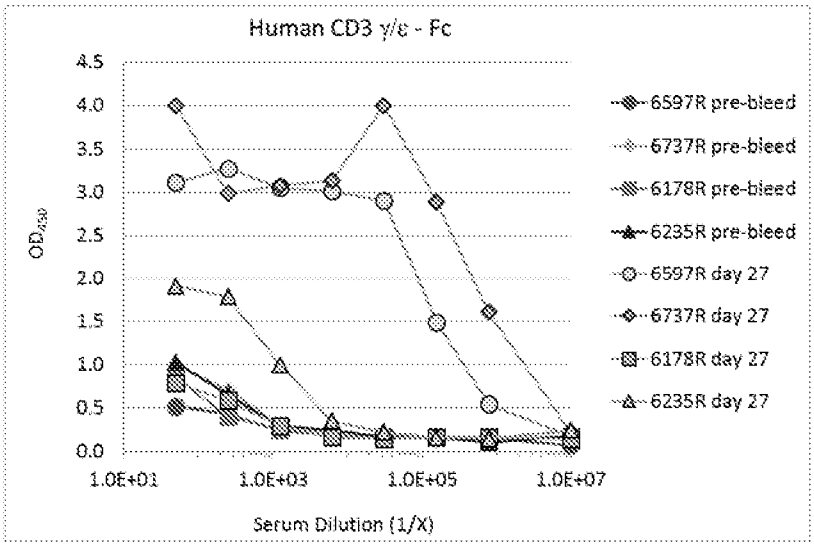


FIGURE 2B

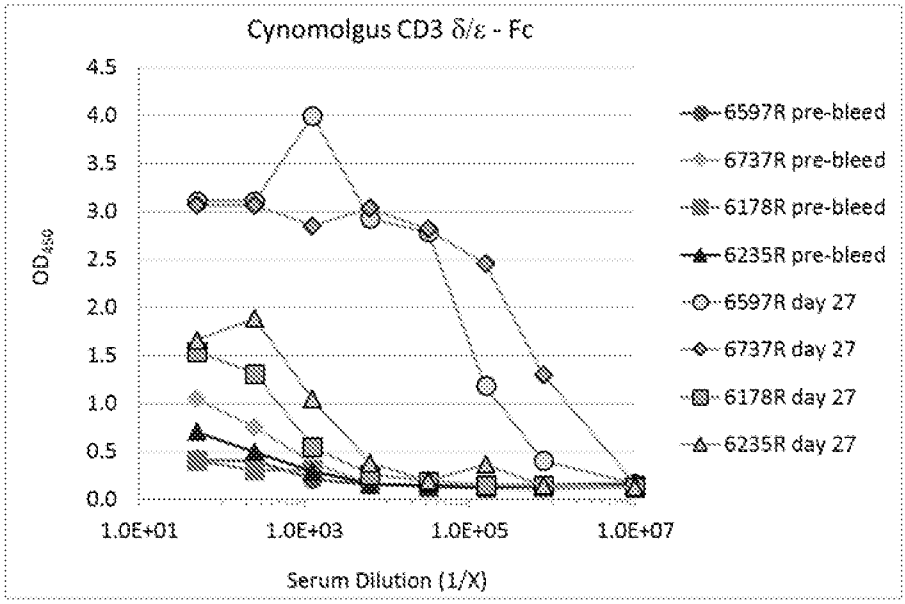


FIGURE 2C

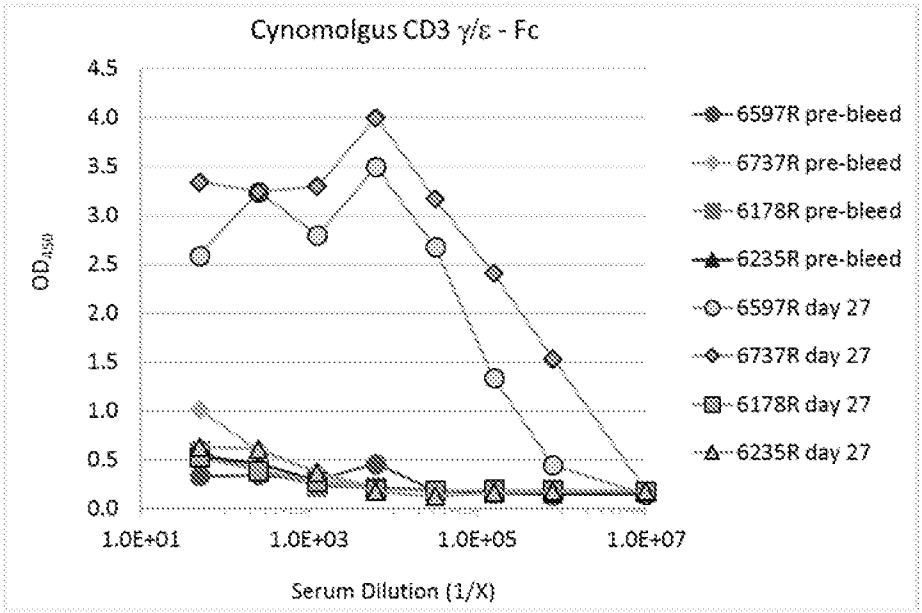


FIGURE 2D

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 3/8

FIGURE 3 provides a summary of screening BCC supernatants for rabbit IgG binding human and cynomolgus T cells by FACS and activation of human CD3+ T cells

		Antibody in B cell culture well			
	BCC well ID	T activate IL-2 (OD)	Parental HEK 293's (MFI)	Cyno T cell line (MFI)	Human CD3+ T cells (MFI)
1	284A10	4.0	15	615	413
2	284C10	4.0	16	432	402
3	293G4	4.0	22	219	359
4	299C5	3.5	15	565	527
5	282A6	3.5	25	266	454
6	299F6	3.4	18	1109	847
7	281E8	3.1	18	183	299
8	295E9	3.1	17	549	478
9	313H9	2.9	35	254	307
10	295A8	2.9	16	431	540
11	290G5	2.8	23	192	255
12	292D3	2.7	35	290	285
13	286E5	2.3	24	33	25
14	289A4	2.1	17	43	70
15	290A12	1.9	4	158	75
16	302G8	1.8	22	277	184
17	298E4	1.7	16	264	215
18	299E6	1.7	30	114	170
19	291H9	1.7	14	528	910
20	284C9	0.9	34	20	18
21	283E3	0.8	20	273	527
22	284B1	0.6	28	527	456
23	284A9	0.6	19	25	27

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 4/8

FIGURE 4 shows the binding of example chimeric rabbit/human IgG antibodies to Cynomolgus CD3 Delta, Gamma, or Epsilon Fc fusion proteins and human T cell line Jurkat **(nd = not done)**

		Antibody in B cell culture well				Chimeric Antibody		
	BCC well ID	T activate IL-2 (OD)	Parental HEK 293's (MFI)	Cyno T cell line (MFI)	Human CD3+ T cells (MFI)	Off-rate Cynomolgus δ/ε - Fc	Off-rate Cynomolgus γ/ε - Fc	FACS binding Human CD3+ T cell line Jurkat
1	284A10	4.0	15	615	413	3.2E-03	1.3E-03	+++++
2	284C10	4.0	16	432	402	-	-	nd
3	293G4	4.0	22	219	359	-	-	nd
4	299C5	3.5	15	565	527	-	-	nd
5	282A6	3.5	25	266	454	-	-	nd
6	299F6	3.4	18	1109	847	5.2E-04	4.6E-04	+++++
7	281E8	3.1	18	183	299	-	-	nd
8	295E9	3.1	17	549	478	5.2E-03	5.2E-03	+++++
9	313H9	2.9	35	254	307	-	-	nd
10	295A8	2.9	16	431	540	-	-	nd
11	290G5	2.8	23	192	255	-	-	nd
12	292D3	2.7	35	290	285	-	-	nd
13	286E5	2.3	24	33	25	-	-	nd
14	289A4	2.1	17	43	70	-	-	nd
15	290A12	1.9	4	158	75	-	-	nd
16	302G8	1.8	22	277	184	-	-	nd
17	298E4	1.7	16	264	215	-	-	nd
18	299E6	1.7	30	114	170	-	-	nd
19	291H9	1.7	14	528	910	1.8E-02	1.8E-02	-
20	284C9	0.9	34	20	18	nd	nd	nd
21	283E3	0.8	20	273	527	3.5E-03	3.9E-03	+++++
22	284B1	0.6	28	527	456	-	-	nd
23	284A9	0.6	19	25	27	3.3E-03	-	-

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 5/8

FIGURE 5 shows the FACS analysis of example chimeric rabbit/human CD3-specific antibodies binding to the human T cell line Jurkat. 5A: FACS analysis of example chimeric rabbit/human CD3-specific antibody 283E3 binding to the human T cell line Jurkat; 5B FACS analysis of example chimeric rabbit/human CD3-specific antibody 284A10 binding to the human T cell line Jurkat; 5C: FACS analysis of example chimeric rabbit/human CD3-specific antibody 284A9 binding to the human T cell line Jurkat; 5D: FACS analysis of example chimeric rabbit/human CD3-specific antibody 299F6 binding to the human T cell line Jurkat; 5E: FACS analysis of example chimeric rabbit/human CD3-specific antibody 295E9 binding to the human T cell line Jurkat; and 5F: FACS analysis of example chimeric rabbit/human CD3-specific antibody 291H9 binding to the human T cell line Jurkat.

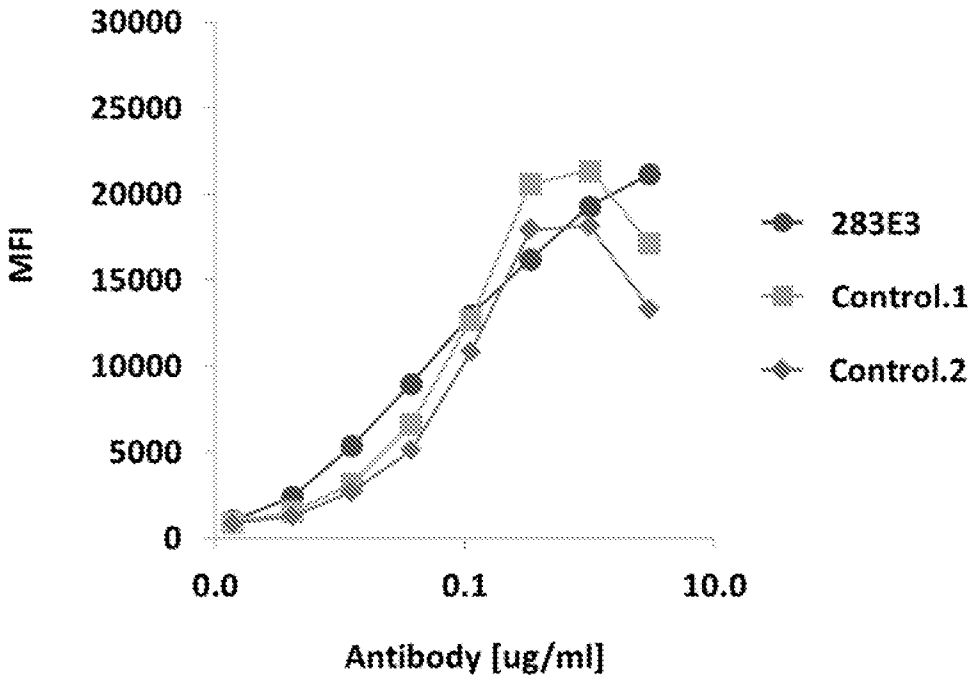


FIGURE 5A

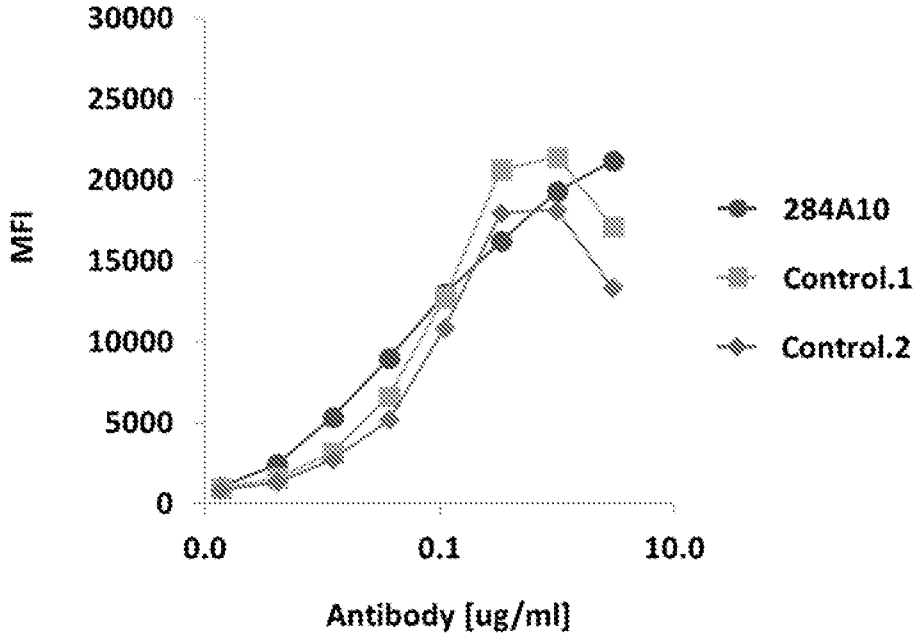


FIGURE 5B

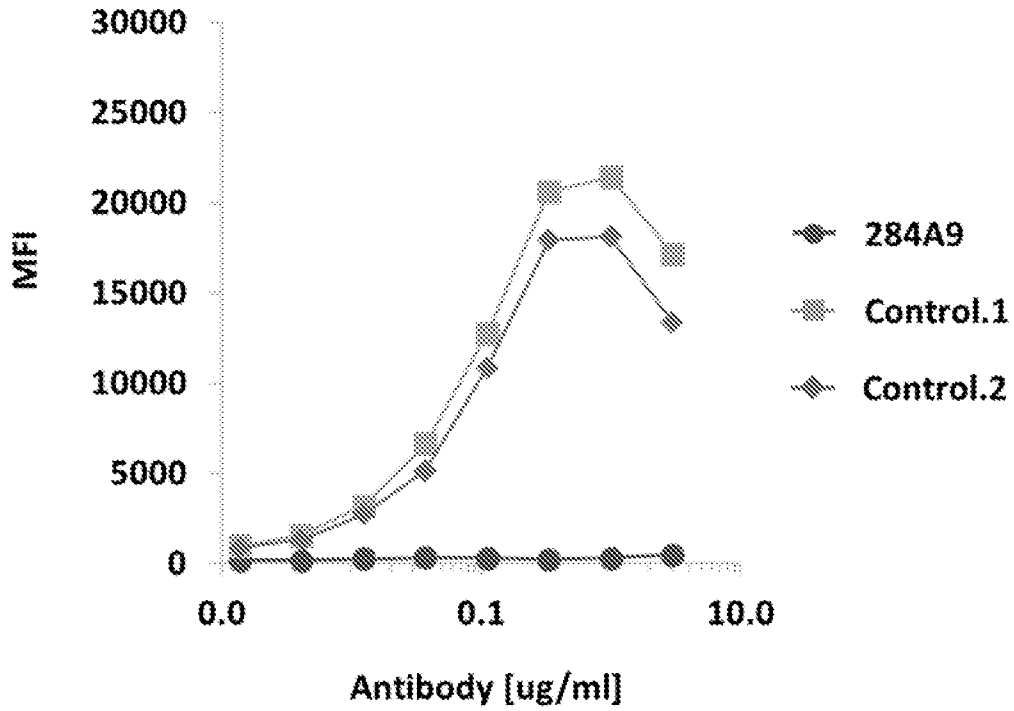


FIGURE 5C

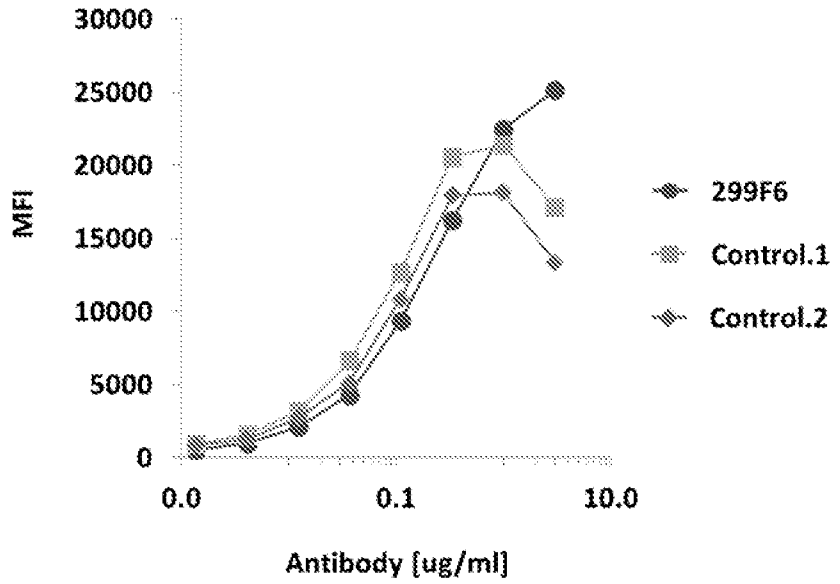

30000
25000
20000
15000
10000
5000
0
MFI
0.0
0.1
10.0
Antibody [ug/ml]
299F6
Control.1
Control.2

FIGURE 5D

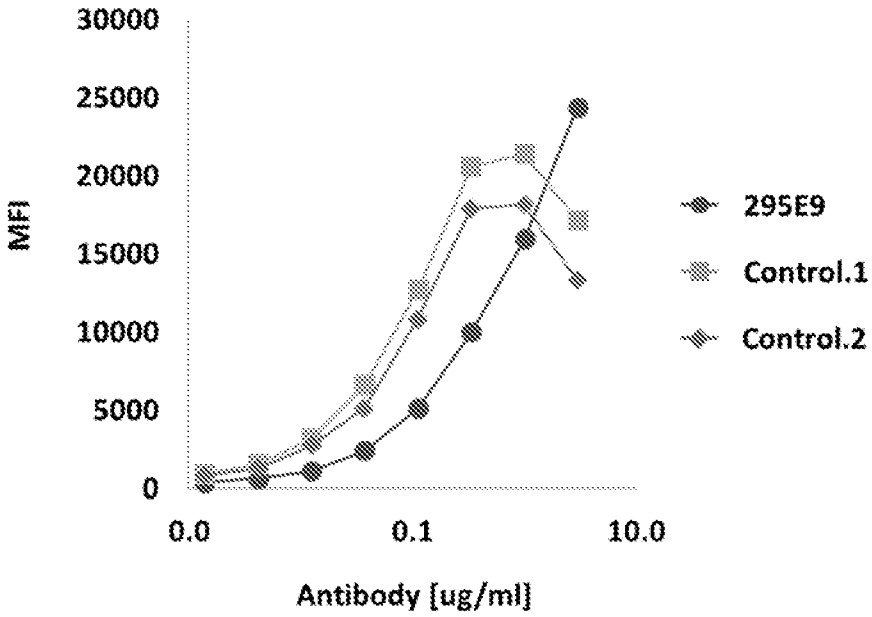

30000
25000
20000
15000
10000
5000
0
MFI
0.0
0.1
10.0
Antibody [ug/ml]
295E9
Control.1
Control.2

FIGURE 5E

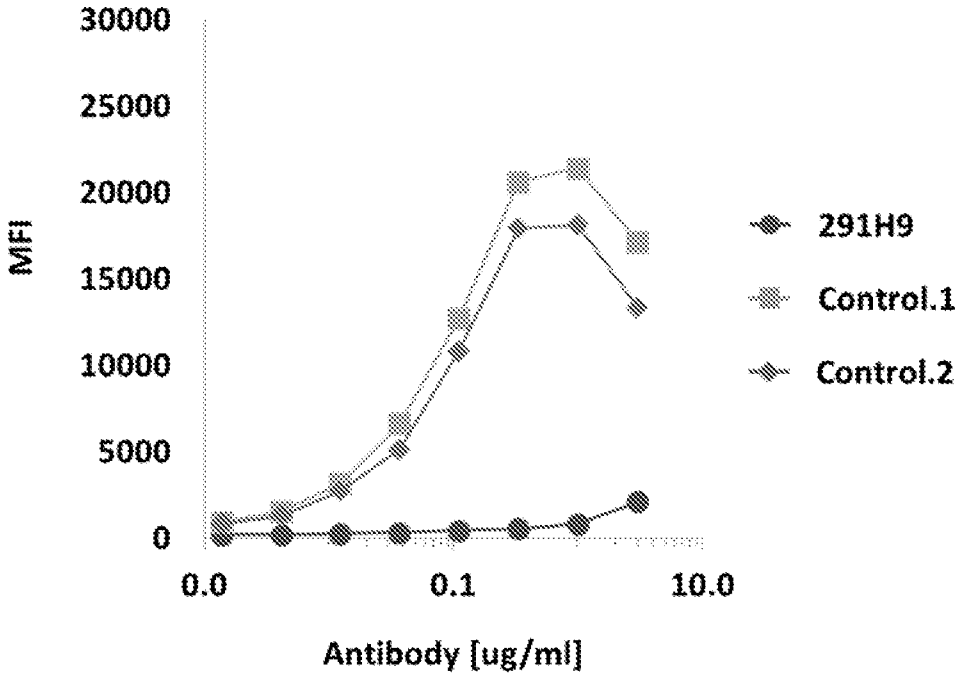


FIGURE 5F

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 6/8

FIGURE 6 shows the FACS analysis of example chimeric rabbit/human CD3-specific antibodies binding to the human T cell line Jurkat. 6A: FACS analysis of example chimeric rabbit/human CD3-specific antibody 496C5 binding to the human T cell line Jurkat; 6B: FACS analysis of example chimeric rabbit/human CD3-specific antibody 480C8 binding to the human T cell line Jurkat; 6C: FACS analysis of example chimeric rabbit/human CD3-specific antibody 480A7 binding to the human T cell line Jurkat; 6D: FACS analysis of example chimeric rabbit/human CD3-specific antibody 485G11 binding to the human T cell line Jurkat; and 6E: FACS analysis of example chimeric rabbit/human CD3-specific antibody 485H6 binding to the human T cell line Jurkat.

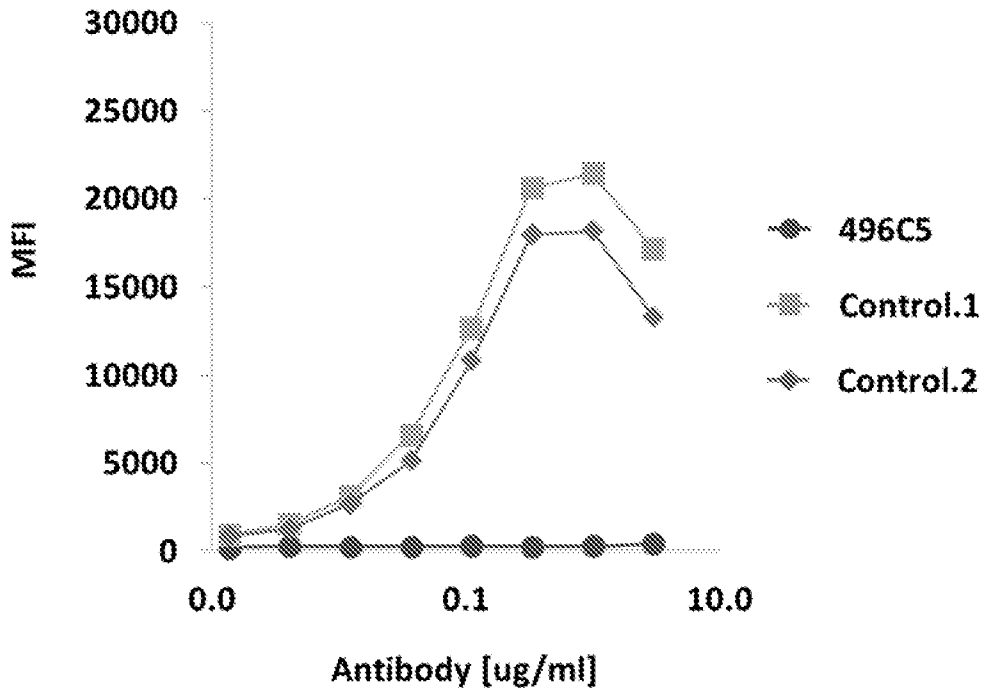


FIGURE 6A

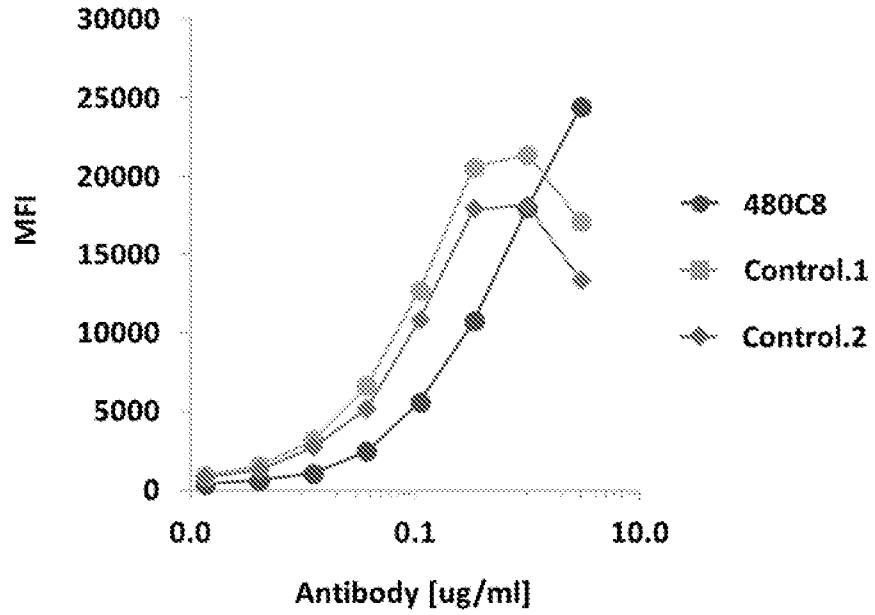


FIGURE 6B

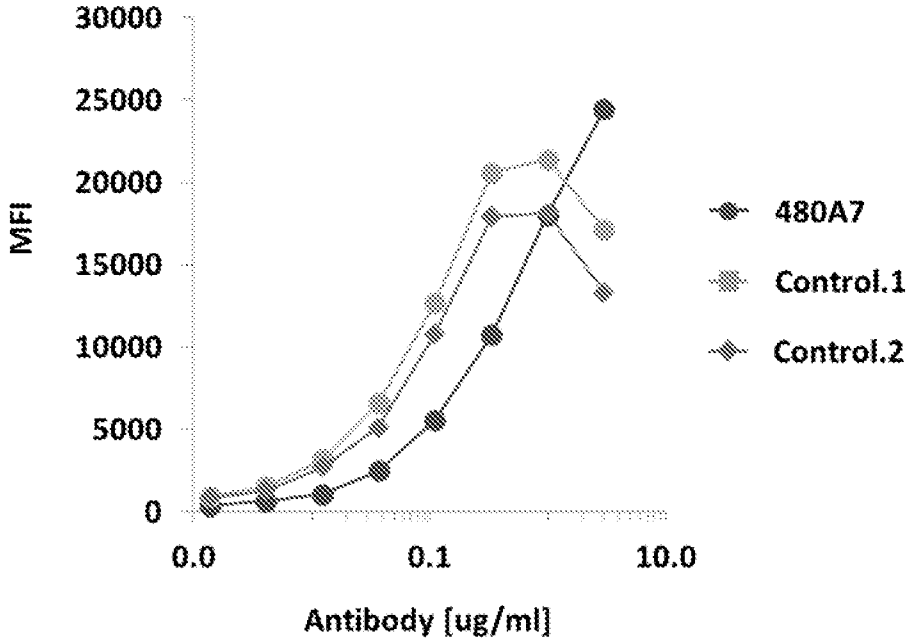


FIGURE 6C

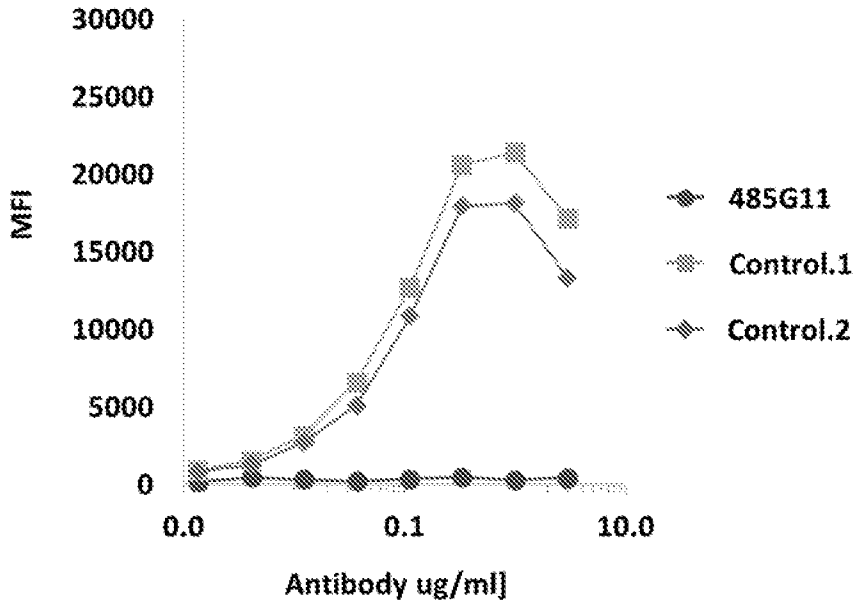


FIGURE 6D

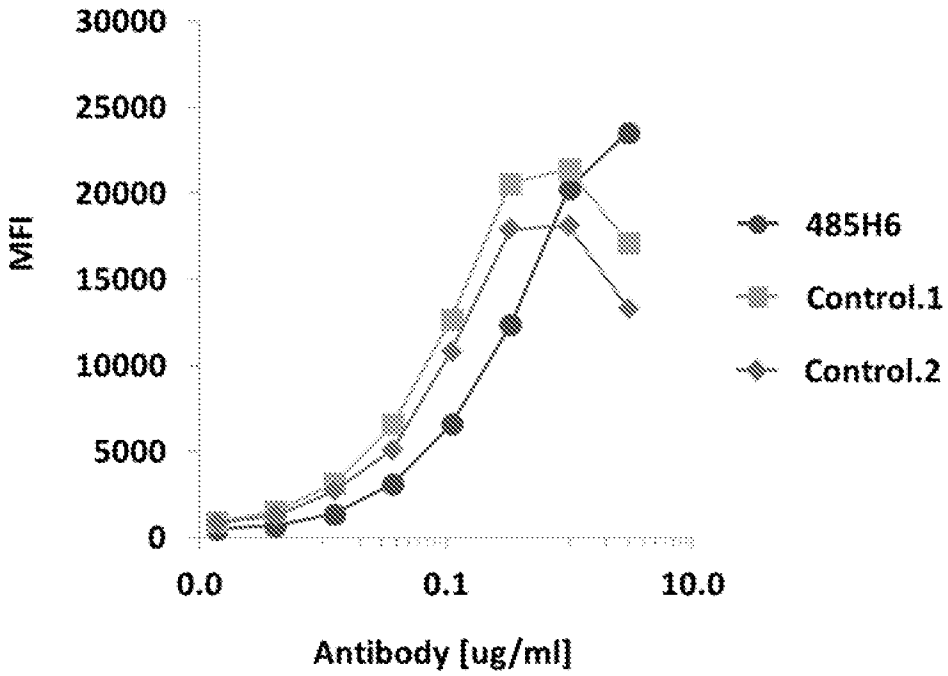
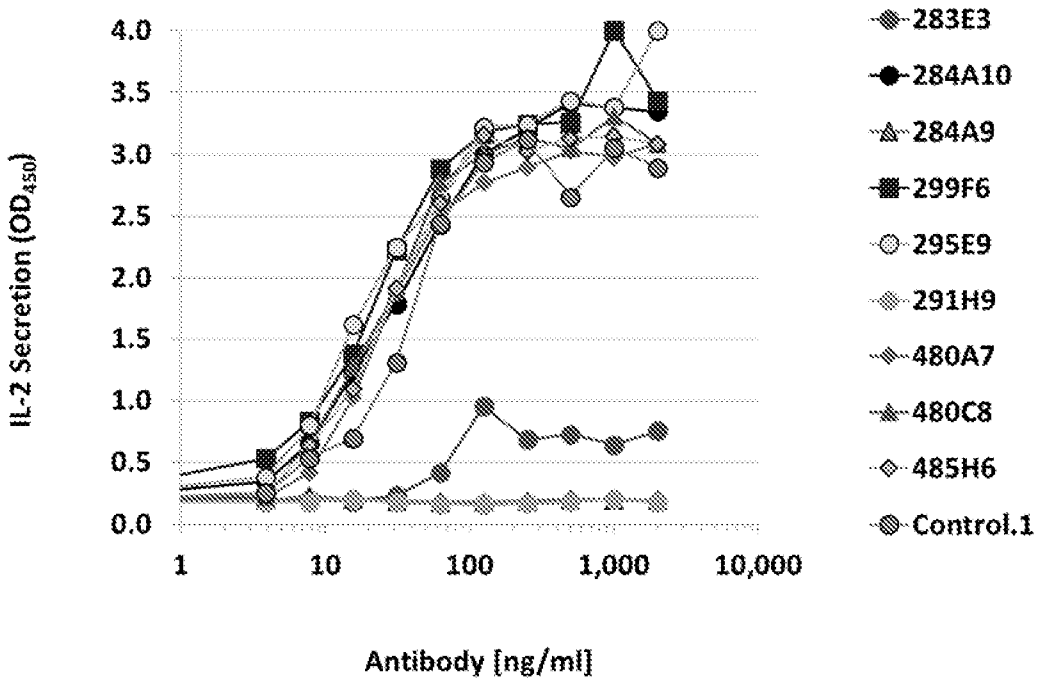


FIGURE 6E

ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 7/8

FIGURE 7 shows the activation of human CD3+ T cells by example chimeric rabbit/human CD3-specific antibodies



ANTI-CD3 ANTIBODIES AND METHODS OF MAKING AND USING THEREOF

FIGURE 8/8

FIGURE 8 shows the binding and off-rate analysis of example chimeric rabbit/human or humanized CD3-specific antibodies

mAb	Format	Antigen	Bivalent kdis (1/s)
284A10	chimeric	Human δ/ε - Fc	5.0E-04
284A10	chimeric	Human δ/γ - Fc	1.3E-03
284A10 1.1	humanized	Human δ/ε - Fc	5.3E-04
299F6	chimeric	Human δ/ε - Fc	4.9E-04
299F6	chimeric	Human δ/γ - Fc	4.5E-04
299F6 1.1	humanized	Human δ/ε - Fc	6.0E-04
480C8	chimeric	Human δ/ε - Fc	4.2E-04
480C8	chimeric	Human δ/γ - Fc	5.4E-04
480C8 1.1	humanized	Human δ/ε - Fc	5.0E-04
295E9	chimeric	Human δ/ε - Fc	5.2E-03
295E9	chimeric	Human δ/γ - Fc	5.2E-03
480A7	chimeric	Human δ/ε - Fc	6.6E-04
480A7	chimeric	Epsilon-Gamma	6.0E-04
485H6	chimeric	Human δ/ε - Fc	5.7E-04
485H6	chimeric	Human δ/γ - Fc	5.3E-04

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 18/39143

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - C07K 16/28, C12N 5/20, G01N 33/577, G01N 33/68, C12R 1/91 (2018.01)
CPC - C07K 16/2809, G01N 33/577, G01N 33/68, G01N 2333/7051

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
See Search History Document

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
See Search History Document

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
See Search History Document

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	KUHN, et al. Therapeutic anti-CD3 monoclonal antibodies: from bench to bedside. Immunotherapy July 2016, Vol 8, No 8, pages 889-906; pg 889, col 1; pg 896, Fig 3 and its lgend	1-10, 13, 15/(1,13), 16-33
A	WO 2016/110584 A1 (BIONTECH AG, et al.) 14 July 2016 (14.07.2016) claim 6, SEQ ID NO:174, 83.6% identity to SEQ ID NO: 4	1-10, 13, 15/(1,13), 16-33
A	FITC anti-human CD3 Antibody, Biolegend, 2012 [online]. [Retrieved on 03 October 2018]. Retrieved from the Internet <URL: https://www.biolegend.com/en-us/global-elements/pdf-popup/fitc-anti-human-cd3-antibody-863?filename=FITC%20anti-human%20CD3%20Antibody.pdf&pdfgen=true >; in entirety	12, 15/(12)
A	US 2013/0302399 A1 (FELDHAUS, et al.) 14 November 2013 (14.11.2013) claim 406, SEQ ID NO: 541, 91.2% identity to SEQ ID NO: 3	12, 15/(12)

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" earlier application or patent but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
12 October 2018	08 JAN 2019

Name and mailing address of the ISA/US	Authorized officer:
Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300	Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

Form PCT/ISA/210 (second sheet) (January 2015)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 18/39143

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:

   a. ☐ forming part of the international application as filed:

      ☐ in the form of an Annex C/ST.25 text file.

      ☐ on paper or in the form of an image file.

   b. ☐ furnished together with the international application under PCT Rule 13*ter*.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.

   c. ☒ furnished subsequent to the international filing date for the purposes of international search only:

      ☒ in the form of an Annex C/ST.25 text file (Rule 13*ter*.1(a)).

      ☒ on paper or in the form of an image file (Rule 13*ter*.1(b) and Administrative Instructions, Section 713).

2. ☒ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

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Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1.

********** See Supplemental Sheet to continue **********

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
1-10, 12, 13, 15/(1,12,13), 16-33, restricted to SEQ ID NO: 4, 3

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

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In Continuation of Box III. Observations where unity of invention is lacking:

Group I+, claims 1-33, directed to an an isolated anti-human CD3 mAb; an isolated nucleic acid encoding thereof, an expression vector; a host cell; a method of producing said antibody; immune-conjugate/ pharmaceutical composition comprising the isolated mAb; a method of comprising administering to the subject an effective amount of the isolated mAb. The anti-hCD3 MAB will be searched to the extent that it encompasses LC of SEQ ID NO: 4, 3 (SEQ ID NO: 3 comprises SEQ ID NO: 4). It is believed that claims 1-10, 12, 13, 15/(1,12,13), 16-33 encompass this first named invention, and thus these claims will be searched without fee to the extent that the anti-hCD3 MAB encompasses LC of SEQ ID NO: 4 or SEQ ID NO: 3. Additional anti-hCD3 MAB(s) will be searched upon the payment of additional fees. Applicants must specify the claims that encompass any additionally elected anti-hCD3 MAB(s). Applicants must further indicate, if applicable, the claims which encompass the first named invention, if different than what was indicated above for this group. Failure to clearly identify how any paid additional invention fees are to be applied to the "+" group(s) will result in only the first claimed invention to be searched. An exemplary election would be an anti-hCD3 MAB comprising HC of SEQ ID NO: 7 or SEQ ID NO: 8 (SEQ ID NO: 7 comprises SEQ ID NO: 8, i.e., claims 1-11, 14, 15/(1,11,14), 16-33.

The inventions listed as Group I+ do not relate to a single special technical feature under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

The inventions special technical feature of some inventions of Group I+ is a LC of a specific amino acid sequence recited therein.
The inventions special technical feature of some inventions of Group I+ is a HC of a specific amino acid sequence recited therein.

The inventions of Group I+ share the technical features of an isolated monoclonal antibody against human CD3. Some inventions of Group I+ share the technical features of an IgG1 HC for an isolated monoclonal antibody against human CD3. Some inventions of Group I+ share the technical features of pharmaceutical composition comprising the isolated mAb or a method of comprising administering to the subject an effective amount of the isolated mAb. However, these shared technical features do not represent an improvement over prior art as being anticipated by a paper titled "Therapeutic anti-CD3 monoclonal antibodies: from bench to bedside" by Kuhn, et al. (Immunotherapy 2016, Vol 8, No 8, pages 889-906) (hereinafter "Kuhn").
Kuhn discloses an isolated monoclonal antibody against human CD3 (pg 889, col 1, " OKT3 (Ortho Kung T3), the first mAb recognizing CD3 surface antigen on human T cells [1]. Marketed under the name muromonab, OKT3 was the first monoclo?nal murine antibody to become available for therapy in humans. In 1986 OKT3 was approved by the US FDA for inhibiting rejection in solid-organ transplantation. This mouse IgG2a is directed against the CD3 epsilon chain of the CD3/TCR complex that characterizes T lymphocytes and has been successfully used to treat allograft rejection in kidney, liver and heart transplantation"; pg 896, Figure 3. New generation of anti-human CD3).

Kuhn discloses an IgG1 HC for an anti-human CD3 MAB (pg 894, col 2, "Teplizumab, also known under the names hOKT3.gamma.1 (Ala-Ala) and MGA031, is a humanized IgG1 antibody that was developed by grafting the complementarity determining region of OKT3 into a human IgG1 backbone. Introduction of two point mutations in its Fc portion decreases binding to FcR").

Kuhn discloses a method of comprising administering to the subject an effective amount of the isolated mAb (pg 896, col 1, "European Phase II/III study giving up to a total of 64 mg of the anti-CD3 mAb otelixizumab over 6 consecutive days reported a long-lasting therapeutic effect..."). As the technical features were known in the art at the time of the invention, they cannot be considered special technical features that would otherwise unify the inventions.

Some inventions of Group I+ share the technical features of a kappa LC for an isolated monoclonal antibody against human CD3. Some inventions of Group I+ share the technical features of a conjugate comprising an isolated monoclonal antibody against human CD3. However, these shared technical features do not represent an improvement over prior art as being anticipated by a publication titled "FITC anti-human CD3 Antibody" by Biolegend (2012) hereinafter Biolegend) [Retrieved from the Internet 03 October 2018: <https://www.biolegend.com/en-us/global-elements/pdf-popup/fitc-anti-human-cd3-antibody-863?filename=FITC%20anti-human%20CD3%20Antibody.pdf&pdfgen=true>].
Biolegend discloses a conjugate of an isolated anti- human CD3 monoclonal antibody comprising a kappa LC (pg 1, FITC anti-human CD3 Antibody, Isotype Mouse IgG1, [kappa]). As the technical features were known in the art at the time of the invention, they cannot be considered special technical features that would otherwise unify the inventions.

Some inventions of Group I+ share the technical features of an isolated nucleic acid encoding an anti-human CD3 MAB, an expression vector comprising thereof; a host cell; or a method of producing said antibody. However, these shared technical features do not represent an improvement over prior art as being anticipated by a paper "Construction and characterization of a humanized anti-human CD3 monoclonal antibody 12F6 with effective immunoregulation functions" by Li, et al. (Immunology 2005, 116(4): 487-498) (hereinafter "Li").
Li discloses an isolated nucleic acid encoding encoding an anti-human CD3 MAB, an expression vector; a host cell; or a method of producing said antibody (pg 488, col 2 to pg 489, col 2, "Cloning of 12F6 heavy and light chain variable region genes RNA was isolated from 12F6 hybridoma cells with TRIzol Reagent ... The heavy and light variable region cDNAs of 12F6 were cloned from hybridoma cells using 5RACE system... Construction of chimeric antibody expression vector... Construction of humanized antibody expression vector The VH of human antibody KOL was chosen as framework for the humanized heavy chain and the VL of human Bence-Jones protein REI was chosen for the humanized light chain. The humanized versions of 12F6 were designed referring to the strategy described by Adair et al.19 The light and heavy chain variable region genes of humanized antibodies were synthesized by overlapping PCR method. The light and heavy chain expression vectors for humanized antibodies were constructed in an identical manner to the chimeric antibody described above... Stable expression and purification of chimeric, humanized, Fc-mutated humanized antibodies Appropriate light and heavy expression vectors were cotransfected into Chinese hamster ovary (CHO)-K1 cells using Lipofectamine 2000 reagent. Stable transfectants were isolated by limiting dilution in the presence of 600 lg/ml G418 and 300 lg/ml Zeocin..."). As the technical features were known in the art at the time of the invention, they cannot be considered special technical features that would otherwise unify the inventions.

Therefore, inventions of Group I+ lack unity under PCT Rule 13 because they do not share the same or corresponding special technical feature.