METHODS AND BIOMARKERS FOR PREDICTING EFFICACY AND EVALUATION OF AN OX40 AGONIST TREATMENT

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the priority benefit of U.S. Provisional Application Serial No. 62/074,612, filed on November 3, 2014, which is incorporated herein by reference in its entirety.

SUBMISSION OF SEQUENCE LISTING ON ASCII TEXT FILE

[0002] The content of the following submission on ASCII text file is incorporated herein by reference in its entirety: a computer readable form (CRF) of the Sequence Listing (file name: 146392028940SEQLIST.TXT, date recorded: November 2, 2015, size: 185 KB).

FIELD

[0003] The present disclosure relates to methods of predicting or monitoring responsiveness to treatment with an OX40 agonist, as well as methods of treating cancer related thereto.

BACKGROUND

[0004] OX40 (also known as CD34, TNFRSF4 and ACT35) is a member of the tumor necrosis factor receptor superfamily. OX40 is not constitutively expressed on naïve T cells, but is induced after engagement of the T cell receptor (TCR). The ligand for OX40, OX40L, is predominantly expressed on antigen presenting cells. OX40 is highly expressed by activated CD4+ T cells, activated CD8+ T cells, memory T cells, and regulatory T cells. OX40 signaling can provide costimulatory signals to CD4 and CD8 T cells, leading to enhanced cell proliferation, survival, effector function and migration. OX40 signaling also enhances memory T cell development and function.

[0005] Regulatory T cells (Treg) cells are highly enriched in tumors and tumor draining lymph nodes derived from multiple cancer indications, including melanoma, NSCLC, renal, ovarian, colon, pancreatic, hepatocellular, and breast cancer. In a subset of these indications, increased intratumoral T reg cell densities are associated with poor patient prognosis, suggesting that these cells play an important role in suppressing antitumor immunity. OX40 positive tumor infiltrating lymphocytes have been described.

[0006] It is clear that there continues to be a need for diagnostic, prognostic, and predictive methods to identify patients that are more likely to benefit from anti-tumor treatments that modulate OX40 activity. The invention described herein meets these needs and provides other benefits.

[0007] All references cited herein, including patent applications, patent publications, and UniProtKB/Swiss-Prot Accession numbers are herein incorporated by reference in their entirety, as if each individual reference were specifically and individually indicated to be incorporated by reference.

BRIEF SUMMARY

[0008] The present disclosure describes methods and biomarkers for predicting efficacy and evaluation of an OX40 agonist treatment, including methods for predicting responsiveness, monitoring pharmacodynamic activity or responsiveness, and methods of treating or delaying progression of cancer.

[0009] The present disclosure provides a method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and (b) classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment.

[0009a] In certain aspects, there is provided a method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and

(b) classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to the OX40 agonist treatment.

[0010] The present disclosure also provides a method for treating or delaying progression of cancer in a subject, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and (b) if the expression level of said one or more marker genes in the sample obtained from the subject is higher than a reference, administering to the subject an effective amount of an OX40 agonist.

[0010a] In certain aspects, the present disclosure provides a use of an OX40 agonist in the manufacture of a medicament for a method for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein said method comprises:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and

(b) wherein the OX40 agonist is to be administered to the subject if the expression level of said one or more marker genes in the tumor sample obtained from the subject is higher than a reference.

[0011] The present disclosure also provides a method for treating or delaying progression of cancer in a subject, comprising administering to the subject an effective amount of an OX40 agonist, wherein a sample comprising leukocytes obtained from the subject has increased expression of one or more marker genes are selected from the group consisting of CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R, as compared with a reference.

[0011a] In certain aspects, the present disclosure provides a use of an OX40 agonist in the manufacture of a medicament for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, wherein the OX40 agonist is to be administered to the subject, and wherein a tumor sample comprising leukocytes obtained from the subject has been detected to possess increased expression of one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R, as compared with a reference.

[0012] In some embodiments, said one or more marker genes are selected from the group consisting of CD8a, CD8b, IFNg, GZMA, GZMB, PRF1, and PDCA1. In some embodiments, said one or more marker genes are selected from the group consisting of H2-d, CTLA4, CXCL9, PTPRC, IL7R, KLRK1, and CXCL1. In some embodiments, said one or more marker genes are selected from the group consisting of CD64, IDO1, and ITGAM.

[0013] The present disclosure also provides a method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and (b)

classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein a decreased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment.

[0013a] In certain aspects, the present disclosure provides a method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and

(b) classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein a decreased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment.

[0014] The present disclosure also provides a method for treating or delaying progression of cancer in a subject, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and (b) if the expression level of said one or more marker genes in the sample obtained from the subject is lower than a reference, administering to the subject an effective amount of an OX40 agonist.

[0014a] In certain aspects, the present disclosure provides a use of an OX40 agonist in the manufacture of a medicament for a method for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein said method comprises:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and

(b) wherein the OX40 agonist is to be administered to the subject if the expression level of said one or more marker genes in the tumor sample obtained from the subject is lower than a reference.

[0015] The present disclosure also provides a method for treating or delaying progression of cancer in a subject, comprising administering to the subject an effective amount of an OX40 agonist, wherein a sample comprising leukocytes obtained from the subject has decreased expression of one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1, as compared with a reference.

[0015a] In certain aspects, the present disclosure provides a use of an OX40 agonist in the manufacture of a medicament for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein the OX40 agonist is to be administered to the subject, and wherein a tumor sample comprising leukocytes obtained from the subject has been detected to possess decreased expression of one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1, as compared with a reference.

[0016] The present disclosure also provides a method for monitoring pharmacodynamic activity of an OX40 agonist treatment, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an OX40 agonist, and wherein said one or more marker genes are selected from the group consisting of ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1, PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and TNFSF4; and (b) determining the treatment as demonstrating pharmacodynamic activity based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates pharmacodynamic activity to the OX40 agonist treatment. In some embodiments, said one or more marker genes are selected from the group consisting of CD3, CD8, IFNg, GZMA, GZMB, PRF1, TNFa, PDCD1, and CD274.

[0016a] In certain aspects, the present disclosure provides a method for monitoring pharmacodynamic activity of an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an anti-OX40 agonist antibody, wherein the subject has cancer, and wherein said one or more marker genes are selected from the group consisting of ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1,

PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and TNFSF4; and

(b) determining the treatment as demonstrating pharmacodynamic activity based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates pharmacodynamic activity to the OX40 agonist treatment.

[0017] The present disclosure also provides a method for monitoring responsiveness of a subject to an OX40 agonist treatment, comprising: (a) measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an OX40 agonist, and wherein said one or more marker genes are selected from the group consisting of BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, H2-aa, H2-d, H2-k, ICOS, IL10, PDCA1, and TNFRSF18; and (b) classifying the subject as responsive or non-responsive to said treatment based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates a responsive subject. In some embodiments, said one or more marker genes are selected from

the group consisting of CD80, CD86, ICOS, H2-aa, and CXCR3. In some embodiments, responsiveness comprises immune activation and/or treatment efficacy.

[0017a] In certain aspects, the present disclosure provides a method for monitoring responsiveness of a subject to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an anti-OX40 agonist antibody, wherein the subject has cancer, and wherein said one or more marker genes are selected from the group consisting of BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, HLA-DQA1, HLA-B, HLA-A, ICOS, IL10, PDCA1, and TNFRSF18; and

(b) classifying the subject as responsive or non-responsive to said treatment based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates a responsive subject. In some embodiments, said one or more marker genes are selected from the group consisting of CD80, CD86, ICOS, H2-aa, and CXCR3. In some embodiments, responsiveness comprises immune activation and/or treatment efficacy.

[0018] In some embodiments, the sample comprising leukocytes is from a tumor sample obtained from the subject. In some embodiments, the sample comprising leukocytes is from a peripheral blood sample obtained from the subject. In some embodiments, the expression level of said one or more marker genes is normalized to the expression level of a reference gene in the sample. In some embodiments, the reference gene is a housekeeping gene. In

some embodiments, the expression level of said one or more marker genes is mRNA expression level. In some embodiments, the mRNA expression level is measured by an assay selected from the group consisting of quantitative PCR, semi-quantitative PCR, nucleotide microarray, RNA-seq, in situ hybridization, and Northern blotting. In some embodiments, the expression level of said one or more marker genes is protein expression level. In some embodiments, the protein expression level is measured by Western blotting, peptide microarray, immunohistochemistry, flow cytometry, or mass spectrometry. In some embodiments, the cancer is selected from the group consisting of colorectal cancer, non-small cell lung cancer, renal cell carcinoma, bladder cancer, ovarian cancer, glioblastoma, neuroblastoma, melanoma, breast carcinoma, gastric cancer, and hepatocellular carcinoma. In some embodiments, the breast carcinoma is triple-negative breast carcinoma. In some embodiments, the OX40 agonist is an agonist anti-human OX40 antibody. In some embodiments, the antibody is a monoclonal antibody. In some embodiments, the antibody is a humanized or human antibody. In some embodiments, the antibody comprises an IgG1 Fc region. In some embodiments, the antibody comprises an IgG4 Fc region. In some embodiments, the antibody comprises an Fc region comprising a mutation that decreases binding to an Fc receptor. In some embodiments, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, 10, 11, 12, 13, or 14; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, 15 or 19; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:7, 22, 23, 24, 25, 26, 27 or 28. In some embodiments, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:7. In some embodiments, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:26. In some embodiments, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2

comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:27. In some embodiments, the antibody is MEDI6469 or MEDI0562. In some embodiments, the OX40 agonist comprises one or more extracellular domains of OX40L. In some embodiments, the OX40 agonist is MEDI6383.

[0019] It is to be understood that one, some, or all of the properties of the various embodiments described above and herein may be combined to form other embodiments of the present invention. These and other aspects of the invention will become apparent to one of skill in the art. These and other embodiments of the invention are further described by the detailed description that follows.

BRIEF DESCRIPTION OF THE DRAWINGS

[0020] **FIGS. 1A & 1B** show the effects of anti-OX40 treatment on T cell function. The graphs depict T cell proliferation (**FIG. 1A**) and IFNγ production (**FIG. 1B**) in response to activation with anti-CD3 and anti-OX40 treatment.

[0021] **FIG. 2** shows the effect of anti-OX40 treatment on Treg activity in an *in vitro* Treg suppression assay, as compared to control treatment.

[0022] **FIGS. 3A-3C** show the effect of anti-OX40 treatment on tumor volume in syngeneic mouse tumor models. Results from the EMT6 breast (**FIG. 3A**), Cloudman melanoma (**FIG. 3B**), and CT26 CRC (**FIG. 3C**) tumor models are shown.

[0023] **FIGS. 4A-4C** show the effect of anti-OX40 treatment on tumor volume in syngeneic mouse tumor models. Results from the MC38 CRC (**FIG. 4A**), 51Blim10 CRC (**FIG. 4B**), and JC breast (**FIG. 4C**) tumor models are shown.

[0024] **FIG. 5** provides a heat-map of gene expression in mouse tumor models (as labeled) prior to anti-OX40 treatment.

[0025] **FIGS. 6A & 6B** demonstrate a dose-dependent effect of anti-OX40 treatment on peripheral blood cells in EMT6 tumor-bearing mice. Shown are the percentages of Tregs (**FIG. 6A**) and CD8 T cells (**FIG. 6B**) in peripheral blood following treatment with the labeled concentration of anti-OX40 antibody.

[0026] **FIGS. 7A & 7B** show the effect of anti-OX40 treatment on numbers of Treg (**FIG. 7A**) and CD8 (**FIG. 7B**) cell infiltrates in EMT6 tumors, compared to control treatment.

[0027] **FIGS. 8A & 8B** show the effect of anti-OX40 treatment on numbers of Treg (**FIG. 8A**) and CD8 (**FIG. 8B**) cell infiltrates in JC tumors, compared to control treatment.

[0028] **FIGS. 9A-9D** show genes that are upregulated following anti-OX40 treatment, compared to control treatment, in both EMT6 and JC tumor models. Shown are relative expression levels of IFNg (**FIG. 9A**), granzyme A (**FIG. 9B**), perforin (**FIG. 9C**), and TNFa (**FIG. 9D**).

[0029] **FIGS. 10A-10D** show genes that are upregulated following anti-OX40 treatment, compared to control treatment, in an EMT6 tumor model. Shown are relative expression levels of H2-aa (**FIG. 10A**), CD86 (**FIG. 10B**), ICOS (**FIG. 10C**), and CXCR3 (**FIG. 10D**).

DETAILED DESCRIPTION

I. General techniques

[0030] The techniques and procedures described or referenced herein are generally well understood and commonly employed using conventional methodology by those skilled in the art, such as, for example, the widely utilized methodologies described in Sambrook et al., *Molecular Cloning: A Laboratory Manual* 3d edition (2001) Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y.; *Current Protocols in Molecular Biology* (F.M. Ausubel, et al. eds., (2003)); the series *Methods in Enzymology* (Academic Press, Inc.): *PCR 2: A Practical Approach* (M.J. MacPherson, B.D. Hames and G.R. Taylor eds. (1995)), Harlow and Lane, eds. (1988) *Antibodies, A Laboratory Manual*, and *Animal Cell Culture* (R.I. Freshney, ed. (1987)); *Oligonucleotide Synthesis* (M.J. Gait, ed., 1984); *Methods in Molecular Biology*, Humana Press; *Cell Biology: A Laboratory Notebook* (J.E. Cellis, ed., 1998) Academic Press; *Animal Cell Culture* (R.I. Freshney), ed., 1987); *Introduction to Cell and Tissue Culture* (J.P. Mather and P.E. Roberts, 1998) Plenum Press; *Cell and Tissue Culture: Laboratory Procedures* (A. Doyle, J.B. Griffiths, and D.G. Newell, eds., 1993-8) J. Wiley and Sons; *Handbook of Experimental Immunology* (D.M. Weir and C.C. Blackwell, eds.); *Gene Transfer Vectors for Mammalian Cells* (J.M. Miller and M.P. Calos, eds., 1987); *PCR: The Polymerase Chain Reaction*, (Mullis et al., eds., 1994); *Current Protocols in Immunology* (J.E. Coligan et al., eds., 1991); *Short Protocols in Molecular Biology* (Wiley and Sons, 1999); *Immunobiology* (C.A. Janeway and P. Travers, 1997); *Antibodies* (P. Finch, 1997); *Antibodies: A Practical Approach* (D. Catty., ed., IRL Press, 1988-1989); *Monoclonal Antibodies: A Practical Approach* (P. Shepherd and C. Dean, eds., Oxford University Press, 2000); *Using Antibodies: A Laboratory Manual* (E. Harlow and D. Lane (Cold Spring Harbor Laboratory Press, 1999); *The Antibodies* (M. Zanetti and J. D. Capra, eds., Harwood

Academic Publishers, 1995); and *Cancer: Principles and Practice of Oncology* (V.T. DeVita et al., eds., J.B. Lippincott Company, 1993).

II. Definitions

[0031] Before describing the invention in detail, it is to be understood that this invention is not limited to particular compositions or biological systems, which can, of course, vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to be limiting.

[0032] As used herein and in the appended claims, the singular forms "a," "or," and "the" include plural referents unless the context clearly dictates otherwise.

[0033] Reference to "about" a value or parameter herein includes (and describes) variations that are directed to that value or parameter per se. For example, description referring to "about X" includes description of "X".

[0034] It is understood that aspects and variations of the invention described herein include "consisting" and/or "consisting essentially of" aspects and variations.

[0035] The term "dysfunction" in the context of immune dysfunction, refers to a state of reduced immune responsiveness to antigenic stimulation.

[0036] The term "dysfunctional", as used herein, also includes refractory or unresponsive to antigen recognition, specifically, impaired capacity to translate antigen recognition into downstream T-cell effector functions, such as proliferation, cytokine production (*e.g.*, gamma interferon) and/or target cell killing.

[0037] "Enhancing T cell function" means to induce, cause or stimulate an effector or memory T cell to have a renewed, sustained or amplified biological function. Examples of enhancing T-cell function include: increased secretion of γ-interferon from $CD8^+$ effector T cells, increased secretion of γ-interferon from CD4+ memory and/or effector T-cells, increased proliferation of CD4+ effector and/or memory T cells, increased proliferation of CD8+ effector T-cells, increased antigen responsiveness (*e.g.*, clearance), relative to such levels before the intervention. In one embodiment, the level of enhancement is at least 50%, alternatively 60%, 70%, 80%, 90%, 100%, 120%, 150%, 200%. The manner of measuring this enhancement is known to one of ordinary skill in the art.

[0038] "Tumor immunity" refers to the process in which tumors evade immune recognition and clearance. Thus, as a therapeutic concept, tumor immunity is "treated" when such evasion is attenuated, and the tumors are recognized and attacked by the immune system. Examples of tumor recognition include tumor binding, tumor shrinkage and tumor clearance.

[0039] "Immunogenicity" refers to the ability of a particular substance to provoke an immune response. Tumors are immunogenic and enhancing tumor immunogenicity aids in the clearance of the tumor cells by the immune response.

[0040] An "acceptor human framework" for the purposes herein is a framework comprising the amino acid sequence of a light chain variable domain (VL) framework or a heavy chain variable domain (VH) framework derived from a human immunoglobulin framework or a human consensus framework, as defined below. An acceptor human framework "derived from" a human immunoglobulin framework or a human consensus framework may comprise the same amino acid sequence thereof, or it may contain amino acid sequence changes. In some embodiments, the number of amino acid changes are 10 or less, 9 or less, 8 or less, 7 or less, 6 or less, 5 or less, 4 or less, 3 or less, or 2 or less. In some embodiments, the VL acceptor human framework is identical in sequence to the VL human immunoglobulin framework sequence or human consensus framework sequence.

[0041] "Affinity" refers to the strength of the sum total of noncovalent interactions between a single binding site of a molecule (e.g., an antibody) and its binding partner (e.g., an antigen). Unless indicated otherwise, as used herein, "binding affinity" refers to intrinsic binding affinity which reflects a 1:1 interaction between members of a binding pair (e.g., antibody and antigen). The affinity of a molecule X for its partner Y can generally be represented by the dissociation constant (Kd). Affinity can be measured by common methods known in the art, including those described herein. Specific illustrative and exemplary embodiments for measuring binding affinity are described in the following.

[0042] An "agonist antibody," as used herein, is an antibody which activates a biological activity of the antigen it binds.

[0043] An "anti-angiogenic agent" refers to a compound which blocks, or interferes with to some degree, the development of blood vessels. An anti-angiogenic agent may, for instance, be a small molecule or antibody that binds to a growth factor or growth factor receptor involved in promoting angiogenesis. In one embodiment, an anti-angiogenic agent is an antibody that binds to vascular endothelial growth factor (VEGF), such as bevacizumab (AVASTIN).

[0044] "Antibody-dependent cell-mediated cytotoxicity" or "ADCC" refers to a form of cytotoxicity in which secreted immunoglobulin bound onto Fc receptors (FcRs) present on certain cytotoxic cells (*e.g.* NK cells, neutrophils, and macrophages) enable these cytotoxic effector cells to bind specifically to an antigen-bearing target cell and subsequently kill the target cell with cytotoxins. The primary cells for mediating ADCC, NK cells, express

FcγRIII only, whereas monocytes express FcγRI, FcγRII, and FcγRIII. FcR expression on hematopoietic cells is summarized in Table 3 on page 464 of Ravetch and Kinet, *Annu. Rev. Immunol* 9:457-92 (1991). To assess ADCC activity of a molecule of interest, an *in vitro* ADCC assay, such as that described in US Patent No. 5,500,362 or 5,821,337 or U.S. Patent No. 6,737,056 (Presta), may be performed. Useful effector cells for such assays include PBMC and NK cells. Alternatively, or additionally, ADCC activity of the molecule of interest may be assessed *in vivo*, *e.g.*, in an animal model such as that disclosed in Clynes *et al. PNAS (USA)* 95:652-656 (1998). An exemplary assay for assessing ADCC activity is provided in the examples herein.

[0045] The terms "anti-OX40 antibody" and "an antibody that binds to OX40" refer to an antibody that is capable of binding OX40 with sufficient affinity such that the antibody is useful as a diagnostic and/or therapeutic agent in targeting OX40. In one embodiment, the extent of binding of an anti-OX40 antibody to an unrelated, non-OX40 protein is less than about 10% of the binding of the antibody to OX40 as measured, e.g., by a radioimmunoassay (RIA). In certain embodiments, an antibody that binds to OX40 has a dissociation constant (Kd) of ≤ 1μM, ≤ 100 nM, ≤ 10 nM, ≤ 1 nM, ≤ 0.1 nM, ≤ 0.01 nM, or ≤ 0.001 nM (e.g. 10^{-8} M or less, e.g. from 10^{-8} M to 10^{-13} M, e.g., from 10^{-9} M to 10^{-13} M). In certain embodiments, an anti-OX40 antibody binds to an epitope of OX40 that is conserved among OX40 from different species.

[0046] As use herein, the term "binds", "specifically binds to" or is "specific for" refers to measurable and reproducible interactions such as binding between a target and an antibody, which is determinative of the presence of the target in the presence of a heterogeneous population of molecules including biological molecules. For example, an antibody that binds to or specifically binds to a target (which can be an epitope) is an antibody that binds this target with greater affinity, avidity, more readily, and/or with greater duration than it binds to other targets. In one embodiment, the extent of binding of an antibody to an unrelated target is less than about 10% of the binding of the antibody to the target as measured, *e.g.,* by a radioimmunoassay (RIA). In certain embodiments, an antibody that specifically binds to a target has a dissociation constant (Kd) of ≤ 1μM, ≤ 100 nM, ≤ 10 nM, ≤ 1 nM, or ≤ 0.1 nM. In certain embodiments, an antibody specifically binds to an epitope on a protein that is conserved among the protein from different species. In another embodiment, specific binding can include, but does not require exclusive binding.

[0047] The term "antibody" herein is used in the broadest sense and encompasses various antibody structures, including but not limited to monoclonal antibodies, polyclonal

antibodies, multispecific antibodies (e.g., bispecific antibodies), and antibody fragments so long as they exhibit the desired antigen-binding activity.

[0048] An "antibody fragment" refers to a molecule other than an intact antibody that comprises a portion of an intact antibody that binds the antigen to which the intact antibody binds. Examples of antibody fragments include but are not limited to Fv, Fab, Fab', Fab'-SH, $F(ab')_2$; diabodies; linear antibodies; single-chain antibody molecules (e.g. scFv); and multispecific antibodies formed from antibody fragments.

[0049] An "antibody that binds to the same epitope" as a reference antibody refers to an antibody that blocks binding of the reference antibody to its antigen in a competition assay by 50% or more, and conversely, the reference antibody blocks binding of the antibody to its antigen in a competition assay by 50% or more. An exemplary competition assay is provided herein.

[0050] The term "binding domain" refers to the region of a polypeptide that binds to another molecule. In the case of an FcR, the binding domain can comprise a portion of a polypeptide chain thereof (*e.g.* the alpha chain thereof) which is responsible for binding an Fc region. One useful binding domain is the extracellular domain of an FcR alpha chain.

[0051] A polypeptide with a variant IgG Fc with "altered" FcR, ADCC or phagocytosis activity is one which has either enhanced or diminished FcR binding activity (e.g, FcγR) and/or ADCC activity and/or phagocytosis activity compared to a parent polypeptide or to a polypeptide comprising a native sequence Fc region.

[0052] The term "OX40," as used herein, refers to any native OX40 from any vertebrate source, including mammals such as primates (e.g. humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses "full-length," unprocessed OX40 as well as any form of OX40 that results from processing in the cell. The term also encompasses naturally occurring variants of OX40, e.g., splice variants or allelic variants. The amino acid sequence of an exemplary human OX40 is shown in SEQ ID NO:1.

[0053] "OX40 activation" refers to activation, of the OX40 receptor. Generally, OX40 activation results in signal transduction.

[0054] The terms "cancer" and "cancerous" refer to or describe the physiological condition in mammals that is typically characterized by unregulated cell growth. Examples of cancer include but are not limited to, carcinoma, lymphoma, blastoma, sarcoma, and leukemia or lymphoid malignancies. More particular examples of such cancers include, but not limited to, squamous cell cancer (*e.g.*, epithelial squamous cell cancer), lung cancer including small-cell lung cancer, non-small cell lung cancer, adenocarcinoma of the lung and squamous

carcinoma of the lung, cancer of the peritoneum, hepatocellular cancer, gastric or stomach cancer including gastrointestinal cancer and gastrointestinal stromal cancer, pancreatic cancer, glioblastoma, cervical cancer, ovarian cancer, liver cancer, bladder cancer, cancer of the urinary tract, hepatoma, breast cancer, colon cancer, rectal cancer, colorectal cancer, endometrial or uterine carcinoma, salivary gland carcinoma, kidney or renal cancer, prostate cancer, vulval cancer, thyroid cancer, hepatic carcinoma, anal carcinoma, penile carcinoma, melanoma, superficial spreading melanoma, lentigo maligna melanoma, acral lentiginous melanomas, nodular melanomas, multiple myeloma and B-cell lymphoma; chronic lymphocytic leukemia (CLL); acute lymphoblastic leukemia (ALL); hairy cell leukemia; chronic myeloblastic leukemia; and post-transplant lymphoproliferative disorder (PTLD), as well as abnormal vascular proliferation associated with phakomatoses, edema (such as that associated with brain tumors), Meigs' syndrome, brain, as well as head and neck cancer, and associated metastases. In certain embodiments, cancers that are amenable to treatment by the antibodies of the invention include breast cancer, colorectal cancer, rectal cancer, non-small cell lung cancer, glioblastoma, non-Hodgkins lymphoma (NHL), renal cell cancer, prostate cancer, liver cancer, pancreatic cancer, soft-tissue sarcoma, kaposi's sarcoma, carcinoid carcinoma, head and neck cancer, ovarian cancer, mesothelioma, and multiple myeloma. In some embodiments, the cancer is selected from: non-small cell lung cancer, glioblastoma, neuroblastoma, melanoma, breast carcinoma (e.g. triple-negative breast cancer), gastric cancer, colorectal cancer (CRC), and hepatocellular carcinoma. Yet, in some embodiments, the cancer is selected from: non-small cell lung cancer, colorectal cancer, glioblastoma and breast carcinoma (e.g. triple-negative breast cancer), including metastatic forms of those cancers.

[0055] The term “tumor” refers to all neoplastic cell growth and proliferation, whether malignant or benign, and all pre-cancerous and cancerous cells and tissues. The terms “cancer,” “cancerous,” “cell proliferative disorder,” “proliferative disorder” and “tumor” are not mutually exclusive as referred to herein.

[0056] The terms “cell proliferative disorder” and “proliferative disorder” refer to disorders that are associated with some degree of abnormal cell proliferation. In one embodiment, the cell proliferative disorder is cancer.

[0057] The term “chimeric” antibody refers to an antibody in which a portion of the heavy and/or light chain is derived from a particular source or species, while the remainder of the heavy and/or light chain is derived from a different source or species.

[0058] The "class" of an antibody refers to the type of constant domain or constant region possessed by its heavy chain. There are five major classes of antibodies: 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, IgG_4, IgA_1, and IgA_2. The heavy chain constant domains that correspond to the different classes of immunoglobulins are called α, δ, ε, γ, and μ, respectively.

[0059] "Complement dependent cytotoxicity" or "CDC" refers to the lysis of a target cell in the presence of complement. Activation of the classical complement pathway is initiated by the binding of the first component of the complement system (C1q) to antibodies (of the appropriate subclass), which are bound to their cognate antigen. To assess complement activation, a CDC assay, *e.g.*, as described in Gazzano-Santoro *et al.*, *J. Immunol. Methods* 202:163 (1996), may be performed. Polypeptide variants with altered Fc region amino acid sequences (polypeptides with a variant Fc region) and increased or decreased C1q binding capability are described, *e.g.*, in US Patent No. 6,194,551 B1 and WO 1999/51642. See also, *e.g.*, Idusogie *et al. J. Immunol.* 164: 4178-4184 (2000).

[0060] The term "cytostatic agent" refers to a compound or composition which arrests growth of a cell either in vitro or in vivo. Thus, a cytostatic agent may be one which significantly reduces the percentage of cells in S phase. Further examples of cytostatic agents include agents that block cell cycle progression by inducing G0/G1 arrest or M-phase arrest. The humanized anti-Her2 antibody trastuzumab (HERCEPTIN®) is an example of a cytostatic agent that induces G0/G1 arrest. Classical M-phase blockers include the vincas (vincristine and vinblastine), taxanes, and topoisomerase II inhibitors such as doxorubicin, epirubicin, daunorubicin, etoposide, and bleomycin. Certain agents that arrest G1 also spill over into S-phase arrest, for example, DNA alkylating agents such as tamoxifen, prednisone, dacarbazine, mechlorethamine, cisplatin, methotrexate, 5-fluorouracil, and ara-C. Further information can be found in Mendelsohn and Israel, eds., *The Molecular Basis of Cancer*, Chapter 1, entitled "Cell cycle regulation, oncogenes, and antineoplastic drugs" by Murakami et al. (W.B. Saunders, Philadelphia, 1995), *e.g.*, p. 13. The taxanes (paclitaxel and docetaxel) are anticancer drugs both derived from the yew tree. Docetaxel (TAXOTERE®, Rhone-Poulenc Rorer), derived from the European yew, is a semisynthetic analogue of paclitaxel (TAXOL®, Bristol-Myers Squibb). Paclitaxel and docetaxel promote the assembly of microtubules from tubulin dimers and stabilize microtubules by preventing depolymerization, which results in the inhibition of mitosis in cells.

[0061] The term "cytotoxic agent" as used herein refers to a substance that inhibits or prevents a cellular function and/or causes cell death or destruction. Cytotoxic agents include,

but are not limited to, radioactive isotopes (e.g., At^{211}, I^{131}, I^{125}, Y^{90}, Re^{186}, Re^{188}, Sm^{153}, Bi^{212}, P^{32}, Pb^{212} and radioactive isotopes of Lu); chemotherapeutic agents or drugs (e.g., methotrexate, adriamicin, vinca alkaloids (vincristine, vinblastine, etoposide), doxorubicin, melphalan, mitomycin C, chlorambucil, daunorubicin or other intercalating agents); growth inhibitory agents; enzymes and fragments thereof such as nucleolytic enzymes; antibiotics; toxins such as small molecule toxins or enzymatically active toxins of bacterial, fungal, plant or animal origin, including fragments and/or variants thereof; and the various antitumor or anticancer agents disclosed below.

[0062] A "depleting anti-OX40 antibody," is an anti-OX40 antibody that kills or depletes OX40-expressing cells. Depletion of OX40 expressing cells can be achieved by various mechanisms, such as antibody-dependent cell-mediated cytotoxicity and/or phagocytosis. Depletion of OX40-expressing cells may be assayed in vitro, and exemplary methods for in vitro ADCC and phagocytosis assays are provided herein. In some embodiments, the OX40-expressing cell is a human CD4+ effector T cell. In some embodiments, the OX40-expressing cell is a transgenic BT474 cell that expresses human OX40.

[0063] "Effector functions" refer to those biological activities attributable to the Fc region of an antibody, which vary with the antibody isotype. Examples of antibody effector functions include: C1q binding and complement dependent cytotoxicity (CDC); Fc receptor binding; antibody-dependent cell-mediated cytotoxicity (ADCC); phagocytosis; down regulation of cell surface receptors (e.g. B cell receptor); and B cell activation.

[0064] An "effective amount" of an agent, e.g., a pharmaceutical formulation, refers to an amount effective, at dosages and for periods of time necessary, to achieve the desired therapeutic or prophylactic result.

[0065] "Fc receptor" or "FcR" describes a receptor that binds to the Fc region of an antibody. In some embodiments, an FcR is a native human FcR. In some embodiments, an FcR is one which binds an IgG antibody (a gamma receptor) and includes receptors of the FcγRI, FcγRII, and FcγRIII subclasses, including allelic variants and alternatively spliced forms of those receptors. FcγRII receptors include FcγRIIA (an "activating receptor") and FcγRIIB (an "inhibiting receptor"), which have similar amino acid sequences that differ primarily in the cytoplasmic domains thereof. Activating receptor FcγRIIA contains an immunoreceptor tyrosine-based activation motif (ITAM) in its cytoplasmic domain. Inhibiting receptor FcγRIIB contains an immunoreceptor tyrosine-based inhibition motif (ITIM) in its cytoplasmic domain. (see, *e.g.*, Daëron, *Annu. Rev. Immunol.* 15:203-234 (1997)). FcRs are reviewed, for example, in Ravetch and Kinet, *Annu. Rev. Immunol* 9:457-

92 (1991); Capel *et al., Immunomethods* 4:25-34 (1994); and de Haas *et al., J. Lab. Clin. Med.* 126:330-41 (1995). Other FcRs, including those to be identified in the future, are encompassed by the term "FcR" herein. The term "Fc receptor" or "FcR" also includes the neonatal receptor, FcRn, which is responsible for the transfer of maternal IgGs to the fetus (Guyer *et al., J. Immunol.* 117:587 (1976) and Kim *et al., J. Immunol.* 24:249 (1994)) and regulation of homeostasis of immunoglobulins. Methods of measuring binding to FcRn are known (see, *e.g.*, Ghetie and Ward., *Immunol. Today* 18(12):592-598 (1997); Ghetie *et al.*, *Nature Biotechnology*, 15(7):637-640 (1997); Hinton *et al.*, *J. Biol. Chem.* 279(8):6213-6216 (2004); WO 2004/92219 (Hinton *et al.*). Binding to human FcRn *in vivo* and serum half life of human FcRn high affinity binding polypeptides can be assayed, *e.g.*, in transgenic mice or transfected human cell lines expressing human FcRn, or in primates to which the polypeptides with a variant Fc region are administered. WO 2000/42072 (Presta) describes antibody variants with improved or diminished binding to FcRs. See also, *e.g.*, Shields *et al. J. Biol. Chem.* 9(2):6591-6604 (2001).

[0066] The term "Fc region" herein is used to define a C-terminal region of an immunoglobulin heavy chain that contains at least a portion of the constant region. The term includes native sequence Fc regions and variant Fc regions. In one embodiment, a human IgG heavy chain Fc region extends from Cys226, or from Pro230, to the carboxyl-terminus of the heavy chain. However, the C-terminal lysine (Lys447) of the Fc region may or may not be present. Unless otherwise specified herein, numbering of amino acid residues in the Fc region or constant region is according to the EU numbering system, also called the EU index, as described in Kabat et al., *Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, MD, 1991.

[0067] A "functional Fc region" possesses an "effector function" of a native sequence Fc region. Exemplary "effector functions" include C1q binding; CDC; Fc receptor binding; ADCC; phagocytosis; down regulation of cell surface receptors (e.g. B cell receptor; BCR), etc. Such effector functions generally require the Fc region to be combined with a binding domain (*e.g.*, an antibody variable domain) and can be assessed using various assays as disclosed, for example, in definitions herein.

[0068] "Human effector cells" refer to leukocytes that express one or more FcRs and perform effector functions. In certain embodiments, the cells express at least FcγRIII and perform ADCC effector function(s). Examples of human leukocytes which mediate ADCC include peripheral blood mononuclear cells (PBMC), natural killer (NK) cells, monocytes,

cytotoxic T cells, and neutrophils. The effector cells may be isolated from a native source, e.g., from blood.

[0069] "Framework" or "FR" refers to variable domain residues other than hypervariable region (HVR) residues. The FR of a variable domain generally consists of four FR domains: FR1, FR2, FR3, and FR4. Accordingly, the HVR and FR sequences generally appear in the following sequence in VH (or VL): FR1-H1(L1)-FR2-H2(L2)-FR3-H3(L3)-FR4.

[0070] The terms "full length antibody," "intact antibody," and "whole antibody" are used herein interchangeably to refer to an antibody having a structure substantially similar to a native antibody structure or having heavy chains that contain an Fc region as defined herein.

[0071] The terms "host cell," "host cell line," and "host cell culture" are used interchangeably and refer to cells into which exogenous nucleic acid has been introduced, including the progeny of such cells. Host cells include "transformants" and "transformed cells," which include the primary transformed cell and progeny derived therefrom without regard to the number of passages. Progeny may not be completely identical in nucleic acid content to a parent cell, but may contain mutations. Mutant progeny that have the same function or biological activity as screened or selected for in the originally transformed cell are included herein.

[0072] A "human antibody" is one which possesses an amino acid sequence which corresponds to that of an antibody produced by a human or a human cell or derived from a non-human source that utilizes human antibody repertoires or other human antibody-encoding sequences. This definition of a human antibody specifically excludes a humanized antibody comprising non-human antigen-binding residues.

[0073] A "human consensus framework" is a framework which represents the most commonly occurring amino acid residues in a selection of human immunoglobulin VL or VH framework sequences. Generally, the selection of human immunoglobulin VL or VH sequences is from a subgroup of variable domain sequences. Generally, the subgroup of sequences is a subgroup as in Kabat et al., *Sequences of Proteins of Immunological Interest*, Fifth Edition, NIH Publication 91-3242, Bethesda MD (1991), vols. 1-3. In one embodiment, for the VL, the subgroup is subgroup kappa I as in Kabat et al., *supra*. In one embodiment, for the VH, the subgroup is subgroup III as in Kabat et al., *supra*.

[0074] A "humanized" antibody refers to a chimeric antibody comprising amino acid residues from non-human HVRs and amino acid residues from human FRs. In certain embodiments, a humanized antibody will comprise substantially all of at least one, and typically two, variable domains, in which all or substantially all of the HVRs (e.g., CDRs)

correspond to those of a non-human antibody, and all or substantially all of the FRs correspond to those of a human antibody. A humanized antibody optionally may comprise at least a portion of an antibody constant region derived from a human antibody. A "humanized form" of an antibody, e.g., a non-human antibody, refers to an antibody that has undergone humanization.

[0075] The term "hypervariable region" or "HVR" as used herein refers to each of the regions of an antibody variable domain which are hypervariable in sequence ("complementarity determining regions" or "CDRs") and/or form structurally defined loops ("hypervariable loops") and/or contain the antigen-contacting residues ("antigen contacts"). Generally, antibodies comprise six HVRs: three in the VH (H1, H2, H3), and three in the VL (L1, L2, L3). Exemplary HVRs herein include:

(a) hypervariable loops occurring at amino acid residues 26-32 (L1), 50-52 (L2), 91-96 (L3), 26-32 (H1), 53-55 (H2), and 96-101 (H3) (Chothia and Lesk, *J. Mol. Biol.* 196:901-917 (1987));

(b) CDRs occurring at amino acid residues 24-34 (L1), 50-56 (L2), 89-97 (L3), 31-35b (H1), 50-65 (H2), and 95-102 (H3) (Kabat et al., *Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, MD (1991));

(c) antigen contacts occurring at amino acid residues 27c-36 (L1), 46-55 (L2), 89-96 (L3), 30-35b (H1), 47-58 (H2), and 93-101 (H3) (MacCallum et al. *J. Mol. Biol.* 262: 732-745 (1996)); and

(d) combinations of (a), (b), and/or (c), including HVR amino acid residues 46-56 (L2), 47-56 (L2), 48-56 (L2), 49-56 (L2), 26-35 (H1), 26-35b (H1), 49-65 (H2), 93-102 (H3), and 94-102 (H3).

[0076] Unless otherwise indicated, HVR residues and other residues in the variable domain (e.g., FR residues) are numbered herein according to Kabat et al., *supra*.

[0077] An "immunoconjugate" is an antibody conjugated to one or more heterologous molecule(s), including but not limited to a cytotoxic agent.

[0078] An "individual" or "subject" is a mammal. Mammals include, but are not limited to, domesticated animals (e.g., cows, sheep, cats, dogs, and horses), primates (e.g., humans and non-human primates such as monkeys), rabbits, and rodents (e.g., mice and rats). In certain embodiments, the individual or subject is a human.

[0079] "Promoting cell growth or proliferation" means increasing a cell's growth or proliferation by at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or 100%.

[0080] An "isolated" antibody is one which has been separated from a component of its natural environment. In some embodiments, an antibody is purified to greater than 95% or 99% purity as determined by, for example, electrophoretic (e.g., SDS-PAGE, isoelectric focusing (IEF), capillary electrophoresis) or chromatographic (e.g., ion exchange or reverse phase HPLC). For review of methods for assessment of antibody purity, see, e.g., Flatman et al., *J. Chromatogr. B* 848:79-87 (2007).

[0081] An "isolated" nucleic acid refers to a nucleic acid molecule that has been separated from a component of its natural environment. An isolated nucleic acid includes a nucleic acid molecule contained in cells that ordinarily contain the nucleic acid molecule, but the nucleic acid molecule is present extrachromosomally or at a chromosomal location that is different from its natural chromosomal location.

[0082] "Isolated nucleic acid encoding an anti-OX40 antibody" refers to one or more nucleic acid molecules encoding antibody heavy and light chains (or fragments thereof), including such nucleic acid molecule(s) in a single vector or separate vectors, and such nucleic acid molecule(s) present at one or more locations in a host cell.

[0083] 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 and/or bind the same epitope, except for possible variant antibodies, e.g., containing naturally occurring mutations or arising during production of a monoclonal antibody preparation, such variants generally being present in minor amounts. In contrast to polyclonal antibody preparations, which typically include different antibodies directed against different determinants (epitopes), each monoclonal antibody of a monoclonal antibody preparation is directed against a single determinant on an antigen. Thus, the modifier "monoclonal" indicates the character of the antibody as being obtained from a substantially homogeneous population of antibodies, and is not to be construed as requiring production of the antibody by any particular method. For example, the monoclonal antibodies to be used in accordance with the present invention may be made by a variety of techniques, including but not limited to the hybridoma method, recombinant DNA methods, phage-display methods, and methods utilizing transgenic animals containing all or part of the human immunoglobulin loci, such methods and other exemplary methods for making monoclonal antibodies being described herein.

[0084] A "naked antibody" refers to an antibody that is not conjugated to a heterologous moiety (e.g., a cytotoxic moiety) or radiolabel. The naked antibody may be present in a pharmaceutical formulation.

[0085] "Native antibodies" refer to naturally occurring immunoglobulin molecules with varying structures. For example, native IgG antibodies are heterotetrameric glycoproteins of about 150,000 daltons, composed of two identical light chains and two identical heavy chains that are disulfide-bonded. From N- to C-terminus, each heavy chain has a variable region (VH), also called a variable heavy domain or a heavy chain variable domain, followed by three constant domains (CH1, CH2, and CH3). Similarly, from N- to C-terminus, each light chain has a variable region (VL), also called a variable light domain or a light chain variable domain, followed by a constant light (CL) domain. The light chain of an antibody may be assigned to one of two types, called kappa (κ) and lambda (λ), based on the amino acid sequence of its constant domain. A "native sequence Fc region" comprises an amino acid sequence identical to the amino acid sequence of an Fc region found in nature. Native sequence human Fc regions include a native sequence human IgG1 Fc region (non-A and A allotypes); native sequence human IgG2 Fc region; native sequence human IgG3 Fc region; and native sequence human IgG4 Fc region as well as naturally occurring variants thereof.

[0086] The term "package insert" is used to refer to instructions customarily included in commercial packages of therapeutic products, that contain information about the indications, usage, dosage, administration, combination therapy, contraindications and/or warnings concerning the use of such therapeutic products.

[0087] "Percent (%) amino acid sequence identity" with respect to a reference polypeptide sequence is defined as the percentage of amino acid residues in a candidate sequence that are identical with the amino acid residues in the reference polypeptide sequence, after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent sequence identity, and not considering any conservative substitutions as part of the sequence identity. Alignment for purposes of determining percent amino acid sequence identity can be achieved in various ways that are within the skill in the art, for instance, using publicly available computer software such as BLAST, BLAST-2, ALIGN or Megalign (DNASTAR) software. Those skilled in the art can determine appropriate parameters for aligning sequences, including any algorithms needed to achieve maximal alignment over the full length of the sequences being compared. For purposes herein, however, % amino acid sequence identity values are generated using the sequence comparison computer program ALIGN-2. The ALIGN-2 sequence comparison computer program was authored by Genentech, Inc., and the

source code has been filed with user documentation in the U.S. Copyright Office, Washington D.C., 20559, where it is registered under U.S. Copyright Registration No. TXU510087. The ALIGN-2 program is publicly available from Genentech, Inc., South San Francisco, California, or may be compiled from the source code. The ALIGN-2 program should be compiled for use on a UNIX operating system, including digital UNIX V4.0D. All sequence comparison parameters are set by the ALIGN-2 program and do not vary.

[0088] In situations where ALIGN-2 is employed for amino acid sequence comparisons, the % amino acid sequence identity of a given amino acid sequence A to, with, or against a given amino acid sequence B (which can alternatively be phrased as a given amino acid sequence A that has or comprises a certain % amino acid sequence identity to, with, or against a given amino acid sequence B) is calculated as follows:

100 times the fraction X/Y

where X is the number of amino acid residues scored as identical matches by the sequence alignment program ALIGN-2 in that program's alignment of A and B, and where Y is the total number of amino acid residues in B. It will be appreciated that where the length of amino acid sequence A is not equal to the length of amino acid sequence B, the % amino acid sequence identity of A to B will not equal the % amino acid sequence identity of B to A. Unless specifically stated otherwise, all % amino acid sequence identity values used herein are obtained as described in the immediately preceding paragraph using the ALIGN-2 computer program.

[0089] The term "pharmaceutical formulation" refers to a preparation which is in such form as to permit the biological activity of an active ingredient contained therein to be effective, and which contains no additional components which are unacceptably toxic to a subject to which the formulation would be administered.

[0090] A "pharmaceutically acceptable carrier" refers to an ingredient in a pharmaceutical formulation, other than an active ingredient, which is nontoxic to a subject. A pharmaceutically acceptable carrier includes, but is not limited to, a buffer, excipient, stabilizer, or preservative.

[0091] As used herein, "treatment" (and grammatical variations thereof such as "treat" or "treating") refers to clinical intervention in an attempt to alter the natural course of the individual being treated, and can be performed either for prophylaxis or during the course of clinical pathology. Desirable effects of treatment include, but are not limited to, preventing occurrence or recurrence of disease, alleviation of symptoms, diminishment of any direct or

indirect pathological consequences of the disease, preventing metastasis, decreasing the rate of disease progression, amelioration or palliation of the disease state, and remission or improved prognosis. In some embodiments, antibodies of the invention are used to delay development of a disease or to slow the progression of a disease.

[0092] The term "tumor" refers to all neoplastic cell growth and proliferation, whether malignant or benign, and all pre-cancerous and cancerous cells and tissues. The terms "cancer," "cancerous," "cell proliferative disorder," "proliferative disorder" and "tumor" are not mutually exclusive as referred to herein.

[0093] The term "variable region" or "variable domain" refers to the domain of an antibody heavy or light chain that is involved in binding the antibody to antigen. The variable domains of the heavy chain and light chain (VH and VL, respectively) of a native antibody generally have similar structures, with each domain comprising four conserved framework regions (FRs) and three hypervariable regions (HVRs). (See, e.g., Kindt et al. *Kuby Immunology*, 6th ed., W.H. Freeman and Co., page 91 (2007).) A single VH or VL domain may be sufficient to confer antigen-binding specificity. Furthermore, antibodies that bind a particular antigen may be isolated using a VH or VL domain from an antibody that binds the antigen to screen a library of complementary VL or VH domains, respectively. See, e.g., Portolano et al., *J. Immunol.* 150:880-887 (1993); Clarkson et al., *Nature* 352:624-628 (1991).

[0094] A "variant Fc region" comprises an amino acid sequence which differs from that of a native sequence Fc region by virtue of at least one amino acid modification, preferably one or more amino acid substitution(s). Preferably, the variant Fc region has at least one amino acid substitution compared to a native sequence Fc region or to the Fc region of a parent polypeptide, e.g. from about one to about ten amino acid substitutions, and preferably from about one to about five amino acid substitutions in a native sequence Fc region or in the Fc region of the parent polypeptide. The variant Fc region herein will preferably possess at least about 80% homology with a native sequence Fc region and/or with an Fc region of a parent polypeptide, and most preferably at least about 90% homology therewith, more preferably at least about 95% homology therewith.

[0095] The term "vector," as used herein, refers to a nucleic acid molecule capable of propagating another nucleic acid to which it is linked. The term includes the vector as a self-replicating nucleic acid structure as well as the vector incorporated into the genome of a host cell into which it has been introduced. Certain vectors are capable of directing the expression of nucleic acids to which they are operatively linked. Such vectors are referred to herein as "expression vectors."

[0096] A "VH subgroup III consensus framework" comprises the consensus sequence obtained from the amino acid sequences in variable heavy subgroup III of Kabat et al. In one embodiment, the VH subgroup III consensus framework amino acid sequence comprises at least a portion or all of each of the following sequences: EVQLVESGGGLVQPGGSLRLSCAAS (SEQ ID NO:214)-H1-WVRQAPGKGLEWV (SEQ ID NO:215)-H2-RFTISRDNSKNTLYLQMNSLRAEDTAVYYC (SEQ ID NO:216)-H3-WGQGTLVTVSS (SEQ ID NO:217).

[0097] A "VL subgroup I consensus framework" comprises the consensus sequence obtained from the amino acid sequences in variable light kappa subgroup I of Kabat et al. In one embodiment, the VH subgroup I consensus framework amino acid sequence comprises at least a portion or all of each of the following sequences: DIQMTQSPSSLSASVGDRVTITC (SEQ ID NO:218)-L1-WYQQKPGKAPKLLIY (SEQ ID NO:219)-L2-GVPSRFSGSGSGTDFTLTISSLQPEDFATYYC (SEQ ID NO:220)-L3-FGQGTKVEIK (SEQ ID NO:221).

[0098] The term "cytotoxic agent" as used herein refers to a substance that inhibits or prevents a cellular function and/or causes cell death or destruction. Cytotoxic agents include, but are not limited to, radioactive isotopes (e.g., At211, I131, I125, Y90, Re186, Re188, Sm153, Bi212, P32, Pb212 and radioactive isotopes of Lu); chemotherapeutic agents; growth inhibitory agents; enzymes and fragments thereof such as nucleolytic enzymes; and toxins such as small molecule toxins or enzymatically active toxins of bacterial, fungal, plant or animal origin, including fragments and/or variants thereof. Exemplary cytotoxic agents can be selected from anti-microtubule agents, platinum coordination complexes, alkylating agents, antibiotic agents, topoisomerase II inhibitors, antimetabolites, topoisomerase I inhibitors, hormones and hormonal analogues, signal transduction pathway inhibitors, non-receptor tyrosine kinase angiogenesis inhibitors, immunotherapeutic agents, proapoptotic agents, inhibitors of LDH-A; inhibitors of fatty acid biosynthesis; cell cycle signalling inhibitors; HDAC inhibitors, proteasome inhibitors; and inhibitors of cancer metabolism.

[0099] In one embodiment the cytotoxic agent is selected from anti-microtubule agents, platinum coordination complexes, alkylating agents, antibiotic agents, topoisomerase II inhibitors, antimetabolites, topoisomerase I inhibitors, hormones and hormonal analogues, signal transduction pathway inhibitors, non-receptor tyrosine kinase angiogenesis inhibitors, immunotherapeutic agents, proapoptotic agents, inhibitors of LDH-A, inhibitors of fatty acid biosynthesis, cell cycle signalling inhibitors, HDAC inhibitors, proteasome inhibitors, and

inhibitors of cancer metabolism. In one embodiment the cytotoxic agent is a taxane. In one embodiment the taxane is paclitaxel or docetaxel. In one embodiment the cytotoxic agent is a platinum agent. In one embodiment the cytotoxic agent is an antagonist of EGFR. In one embodiment the antagonist of EGFR is N-(3-ethynylphenyl)-6,7-bis(2-methoxyethoxy)quinazolin-4-amine (e.g., erlotinib). In one embodiment the cytotoxic agent is a RAF inhibitor. In one embodiment, the RAF inhibitor is a BRAF and/or CRAF inhibitor. In one embodiment the RAF inhibitor is vemurafenib. In one embodiment the cytotoxic agent is a PI3K inhibitor.

[0100] "Chemotherapeutic agent" includes chemical compounds useful in the treatment of cancer. Examples of chemotherapeutic agents include erlotinib (TARCEVA®, Genentech/OSI Pharm.), bortezomib (VELCADE®, Millennium Pharm.), disulfiram, epigallocatechin gallate , salinosporamide A, carfilzomib, 17-AAG (geldanamycin), radicicol, lactate dehydrogenase A (LDH-A), fulvestrant (FASLODEX®, AstraZeneca), sunitib (SUTENT®, Pfizer/Sugen), letrozole (FEMARA®, Novartis), imatinib mesylate (GLEEVEC®, Novartis), finasunate (VATALANIB®, Novartis), oxaliplatin (ELOXATIN®, Sanofi), 5-FU (5-fluorouracil), leucovorin, Rapamycin (Sirolimus, RAPAMUNE®, Wyeth), Lapatinib (TYKERB®, GSK572016, Glaxo Smith Kline), Lonafamib (SCH 66336), sorafenib (NEXAVAR®, Bayer Labs), gefitinib (IRESSA®, AstraZeneca), AG1478, alkylating agents such as thiotepa and CYTOXAN® cyclosphosphamide; alkyl sulfonates such as busulfan, improsulfan and piposulfan; aziridines such as benzodopa, carboquone, meturedopa, and uredopa; ethylenimines and methylamelamines including altretamine, triethylenemelamine, triethylenephosphoramide, triethylenethiophosphoramide and trimethylomelamine; acetogenins (especially bullatacin and bullatacinone); a camptothecin (including topotecan and irinotecan); bryostatin; callystatin; CC-1065 (including its adozelesin, carzelesin and bizelesin synthetic analogs); cryptophycins (particularly cryptophycin 1 and cryptophycin 8); adrenocorticosteroids (including prednisone and prednisolone); cyproterone acetate; 5α-reductases including finasteride and dutasteride); vorinostat, romidepsin, panobinostat, valproic acid, mocetinostat dolastatin; aldesleukin, talc duocarmycin (including the synthetic analogs, KW-2189 and CB1-TM1); eleutherobin; pancratistatin; a sarcodictyin; spongistatin; nitrogen mustards such as chlorambucil, chlomaphazine, chlorophosphamide, estramustine, ifosfamide, mechlorethamine, mechlorethamine oxide hydrochloride, melphalan, novembichin, phenesterine, prednimustine, trofosfamide, uracil mustard; nitrosoureas such as carmustine, chlorozotocin, fotemustine, lomustine, nimustine, and ranimnustine; antibiotics such as the enediyne

antibiotics (e.g., calicheamicin, especially calicheamicin γ1I and calicheamicin ω1I (Angew Chem. Intl. Ed. Engl. 1994 33:183-186); dynemicin, including dynemicin A; bisphosphonates, such as clodronate; an esperamicin; as well as neocarzinostatin chromophore and related chromoprotein enediyne antibiotic chromophores), aclacinomysins, actinomycin, authramycin, azaserine, bleomycins, cactinomycin, carabicin, caminomycin, carzinophilin, chromomycinis, dactinomycin, daunorubicin, detorubicin, 6-diazo-5-oxo-L-norleucine, ADRIAMYCIN® (doxorubicin), morpholino-doxorubicin, cyanomorpholino-doxorubicin, 2-pyrrolino-doxorubicin and deoxydoxorubicin), epirubicin, esorubicin, idarubicin, marcellomycin, mitomycins such as mitomycin C, mycophenolic acid, nogalamycin, olivomycins, peplomycin, porfiromycin, puromycin, quelamycin, rodorubicin, streptonigrin, streptozocin, tubercidin, ubenimex, zinostatin, zorubicin; anti-metabolites such as methotrexate and 5-fluorouracil (5-FU); folic acid analogs such as denopterin, methotrexate, pteropterin, trimetrexate; purine analogs such as fludarabine, 6-mercaptopurine, thiamiprine, thioguanine; pyrimidine analogs such as ancitabine, azacitidine, 6-azauridine, carmofur, cytarabine, dideoxyuridine, doxifluridine, enocitabine, floxuridine; androgens such as calusterone, dromostanolone propionate, epitiostanol, mepitiostane, testolactone; anti-adrenals such as aminoglutethimide, mitotane, trilostane; folic acid replenisher such as frolinic acid; aceglatone; aldophosphamide glycoside; aminolevulinic acid; eniluracil; amsacrine; bestrabucil; bisantrene; edatraxate; defofamine; demecolcine; diaziquone; elfomithine; elliptinium acetate; an epothilone; etoglucid; gallium nitrate; hydroxyurea; lentinan; lonidainine; maytansinoids such as maytansine and ansamitocins; mitoguazone; mitoxantrone; mopidamnol; nitraerine; pentostatin; phenamet; pirarubicin; losoxantrone; podophyllinic acid; 2-ethylhydrazide; procarbazine; PSK® polysaccharide complex (JHS Natural Products, Eugene, Oreg.); razoxane; rhizoxin; sizofuran; spirogermanium; tenuazonic acid; triaziquone; 2,2',2"-trichlorotriethylamine; trichothecenes (especially T-2 toxin, verracurin A, roridin A and anguidine); urethan; vindesine; dacarbazine; mannomustine; mitobronitol; mitolactol; pipobroman; gacytosine; arabinoside ("Ara-C"); cyclophosphamide; thiotepa; taxoids, e.g., TAXOL (paclitaxel; Bristol-Myers Squibb Oncology, Princeton, N.J.), ABRAXANE® (Cremophor-free), albumin-engineered nanoparticle formulations of paclitaxel (American Pharmaceutical Partners, Schaumberg, Ill.), and TAXOTERE® (docetaxel, doxetaxel; Sanofi-Aventis); chloranmbucil; GEMZAR® (gemcitabine); 6-thioguanine; mercaptopurine; methotrexate; platinum analogs such as cisplatin and carboplatin; vinblastine; etoposide (VP-16); ifosfamide; mitoxantrone; vincristine; NAVELBINE® (vinorelbine); novantrone; teniposide; edatrexate; daunomycin;

aminopterin; capecitabine (XELODA®); ibandronate; CPT-11; topoisomerase inhibitor RFS 2000; difluoromethylornithine (DMFO); retinoids such as retinoic acid; and pharmaceutically acceptable salts, acids and derivatives of any of the above.

[0101] Chemotherapeutic agent also includes (i) anti-hormonal agents that act to regulate or inhibit hormone action on tumors such as anti-estrogens and selective estrogen receptor modulators (SERMs), including, for example, tamoxifen (including NOLVADEX®; tamoxifen citrate), raloxifene, droloxifene, iodoxyfene , 4-hydroxytamoxifen, trioxifene, keoxifene, LY117018, onapristone, and FARESTON® (toremifine citrate); (ii) aromatase inhibitors that inhibit the enzyme aromatase, which regulates estrogen production in the adrenal glands, such as, for example, 4(5)-imidazoles, aminoglutethimide, MEGASE® (megestrol acetate), AROMASIN® (exemestane; Pfizer), formestanie, fadrozole, RIVISOR® (vorozole), FEMARA® (letrozole; Novartis), and ARIMIDEX® (anastrozole; AstraZeneca); (iii) anti-androgens such as flutamide, nilutamide, bicalutamide, leuprolide and goserelin; buserelin, tripterelin, medroxyprogesterone acetate, diethylstilbestrol, premarin, fluoxymesterone, all transretionic acid, fenretinide, as well as troxacitabine (a 1,3-dioxolane nucleoside cytosine analog); (iv) protein kinase inhibitors; (v) lipid kinase inhibitors; (vi) antisense oligonucleotides, particularly those which inhibit expression of genes in signaling pathways implicated in aberrant cell proliferation, such as, for example, PKC-alpha, Ralf and H-Ras; (vii) ribozymes such as VEGF expression inhibitors (e.g., ANGIOZYME®) and HER2 expression inhibitors; (viii) vaccines such as gene therapy vaccines, for example, ALLOVECTIN®, LEUVECTIN®, and VAXID®; PROLEUKIN®, rIL-2; a topoisomerase 1 inhibitor such as LURTOTECAN®; ABARELIX® rmRH; and (ix) pharmaceutically acceptable salts, acids and derivatives of any of the above.

[0102] Chemotherapeutic agent also includes antibodies such as alemtuzumab (Campath), bevacizumab (AVASTIN®, Genentech); cetuximab (ERBITUX®, Imclone); panitumumab (VECTIBIX®, Amgen), rituximab (RITUXAN®, Genentech/Biogen Idec), pertuzumab (OMNITARG®, 2C4, Genentech), trastuzumab (HERCEPTIN®, Genentech), tositumomab (Bexxar, Corixia), and the antibody drug conjugate, gemtuzumab ozogamicin (MYLOTARG®, Wyeth). Additional humanized monoclonal antibodies with therapeutic potential as agents in combination with the compounds of the invention include: apolizumab, aselizumab, atlizumab, bapineuzumab, bivatuzumab mertansine, cantuzumab mertansine, cedelizumab, certolizumab pegol, cidfusituzumab, cidtuzumab, daclizumab, eculizumab, efalizumab, epratuzumab, erlizumab, felvizumab, fontolizumab, gemtuzumab ozogamicin, inotuzumab ozogamicin, ipilimumab, labetuzumab, lintuzumab, matuzumab, mepolizumab,

motavizumab, motovizumab, natalizumab, nimotuzumab, nolovizumab, numavizumab, ocrelizumab, omalizumab, palivizumab, pascolizumab, pecfusituzumab, pectuzumab, pexelizumab, ralivizumab, ranibizumab, reslivizumab, reslizumab, resyvizumab, rovelizumab, ruplizumab, sibrotuzumab, siplizumab, sontuzumab, tacatuzumab tetraxetan, tadocizumab, talizumab, tefibazumab, tocilizumab, toralizumab, tucotuzumab celmoleukin, tucusituzumab, umavizumab, urtoxazumab, ustekinumab, visilizumab, and the anti–interleukin-12 (ABT-874/J695, Wyeth Research and Abbott Laboratories) which is a recombinant exclusively human-sequence, full-length IgG1 λ antibody genetically modified to recognize interleukin-12 p40 protein.

[0103] Chemotherapeutic agent also includes "EGFR inhibitors," which refers to compounds that bind to or otherwise interact directly with EGFR and prevent or reduce its signaling activity, and is alternatively referred to as an "EGFR antagonist." Examples of such agents include antibodies and small molecules that bind to EGFR. Examples of antibodies which bind to EGFR include MAb 579 (ATCC CRL HB 8506), MAb 455 (ATCC CRL HB8507), MAb 225 (ATCC CRL 8508), MAb 528 (ATCC CRL 8509) (see, US Patent No. 4,943, 533, Mendelsohn et al.) and variants thereof, such as chimerized 225 (C225 or Cetuximab; ERBUTIX®) and reshaped human 225 (H225) (see, WO 96/40210, Imclone Systems Inc.); IMC-11F8, a fully human, EGFR-targeted antibody (Imclone); antibodies that bind type II mutant EGFR (US Patent No. 5,212,290); humanized and chimeric antibodies that bind EGFR as described in US Patent No. 5,891,996; and human antibodies that bind EGFR, such as ABX-EGF or Panitumumab (see WO98/50433, Abgenix/Amgen); EMD 55900 (Stragliotto et al. Eur. J. Cancer 32A:636-640 (1996)); EMD7200 (matuzumab) a humanized EGFR antibody directed against EGFR that competes with both EGF and TGF-alpha for EGFR binding (EMD/Merck); human EGFR antibody, HuMax-EGFR (GenMab); fully human antibodies known as E1.1, E2.4, E2.5, E6.2, E6.4, E2.11, E6. 3 and E7.6. 3 and described in US 6,235,883; MDX-447 (Medarex Inc); and mAb 806 or humanized mAb 806 (Johns et al., J. Biol. Chem. 279(29):30375-30384 (2004)). The anti-EGFR antibody may be conjugated with a cytotoxic agent, thus generating an immunoconjugate (see, e.g., EP659,439A2, Merck Patent GmbH). EGFR antagonists include small molecules such as compounds described in US Patent Nos: 5,616,582, 5,457,105, 5,475,001, 5,654,307, 5,679,683, 6,084,095, 6,265,410, 6,455,534, 6,521,620, 6,596,726, 6,713,484, 5,770,599, 6,140,332, 5,866,572, 6,399,602, 6,344,459, 6,602,863, 6,391,874, 6,344,455, 5,760,041, 6,002,008, and 5,747,498, as well as the following PCT publications: WO98/14451, WO98/50038, WO99/09016, and WO99/24037. Particular small molecule EGFR antagonists

include OSI-774 (CP-358774, erlotinib, TARCEVA® Genentech/OSI Pharmaceuticals); PD 183805 (CI 1033, 2-propenamide, N-[4-[(3-chloro-4-fluorophenyl)amino]-7-[3-(4-morpholinyl)propoxy]-6-quinazolinyl]-, dihydrochloride, Pfizer Inc.); ZD1839, gefitinib (IRESSA®) 4-(3'-Chloro-4'-fluoroanilino)-7-methoxy-6-(3-morpholinopropoxy)quinazoline, AstraZeneca); ZM 105180 ((6-amino-4-(3-methylphenyl-amino)-quinazoline, Zeneca); BIBX-1382 (N8-(3-chloro-4-fluoro-phenyl)-N2-(1-methyl-piperidin-4-yl)-pyrimido[5,4-d]pyrimidine-2,8-diamine, Boehringer Ingelheim); PKI-166 ((R)-4-[4-[(1-phenylethyl)amino]-1H-pyrrolo[2,3-d]pyrimidin-6-yl]-phenol); (R)-6-(4-hydroxyphenyl)-4-[(1-phenylethyl)amino]-7H-pyrrolo[2,3-d]pyrimidine); CL-387785 (N-[4-[(3-bromophenyl)amino]-6-quinazolinyl]-2-butynamide); EKB-569 (N-[4-[(3-chloro-4-fluorophenyl)amino]-3-cyano-7-ethoxy-6-quinolinyl]-4-(dimethylamino)-2-butenamide) (Wyeth); AG1478 (Pfizer); AG1571 (SU 5271; Pfizer); dual EGFR/HER2 tyrosine kinase inhibitors such as lapatinib (TYKERB®, GSK572016 or N-[3-chloro-4-[(3 fluorophenyl)methoxy]phenyl]-6[5[[[2methylsulfonyl)ethyl]amino]methyl]-2-furanyl]-4-quinazolinamine).

[0104] Chemotherapeutic agents also include "tyrosine kinase inhibitors" including the EGFR-targeted drugs noted in the preceding paragraph; small molecule HER2 tyrosine kinase inhibitor such as TAK165 available from Takeda; CP-724,714, an oral selective inhibitor of the ErbB2 receptor tyrosine kinase (Pfizer and OSI); dual-HER inhibitors such as EKB-569 (available from Wyeth) which preferentially binds EGFR but inhibits both HER2 and EGFR-overexpressing cells; lapatinib (GSK572016; available from Glaxo-SmithKline), an oral HER2 and EGFR tyrosine kinase inhibitor; PKI-166 (available from Novartis); pan-HER inhibitors such as canertinib (CI-1033; Pharmacia); Raf-1 inhibitors such as antisense agent ISIS-5132 available from ISIS Pharmaceuticals which inhibit Raf-1 signaling; non-HER targeted TK inhibitors such as imatinib mesylate (GLEEVEC®, available from Glaxo SmithKline); multi-targeted tyrosine kinase inhibitors such as sunitinib (SUTENT®, available from Pfizer); VEGF receptor tyrosine kinase inhibitors such as vatalanib (PTK787/ZK222584, available from Novartis/Schering AG); MAPK extracellular regulated kinase I inhibitor CI-1040 (available from Pharmacia); quinazolines, such as PD 153035,4-(3-chloroanilino) quinazoline; pyridopyrimidines; pyrimidopyrimidines; pyrrolopyrimidines, such as CGP 59326, CGP 60261 and CGP 62706; pyrazolopyrimidines, 4-(phenylamino)-7H-pyrrolo[2,3-d] pyrimidines; curcumin (diferuloyl methane, 4,5-bis (4-fluoroanilino)phthalimide); tyrphostines containing nitrothiophene moieties; PD-0183805 (Warner-Lamber); antisense molecules (e.g. those that bind to HER-encoding nucleic acid);

quinoxalines (US Patent No. 5,804,396); tryphostins (US Patent No. 5,804,396); ZD6474 (Astra Zeneca); PTK-787 (Novartis/Schering AG); pan-HER inhibitors such as CI-1033 (Pfizer); Affinitac (ISIS 3521; Isis/Lilly); imatinib mesylate (GLEEVEC®); PKI 166 (Novartis); GW2016 (Glaxo SmithKline); CI-1033 (Pfizer); EKB-569 (Wyeth); Semaxinib (Pfizer); ZD6474 (AstraZeneca); PTK-787 (Novartis/Schering AG); INC-1C11 (Imclone), rapamycin (sirolimus, RAPAMUNE®); or as described in any of the following patent publications: US Patent No. 5,804,396; WO 1999/09016 (American Cyanamid); WO 1998/43960 (American Cyanamid); WO 1997/38983 (Warner Lambert); WO 1999/06378 (Warner Lambert); WO 1999/06396 (Warner Lambert); WO 1996/30347 (Pfizer, Inc); WO 1996/33978 (Zeneca); WO 1996/3397 (Zeneca) and WO 1996/33980 (Zeneca).

[0105] Chemotherapeutic agents also include dexamethasone, interferons, colchicine, metoprine, cyclosporine, amphotericin, metronidazole, alemtuzumab, alitretinoin, allopurinol, amifostine, arsenic trioxide, asparaginase, BCG live, bevacuzimab, bexarotene, cladribine, clofarabine, darbepoetin alfa, denileukin, dexrazoxane, epoetin alfa, elotinib, filgrastim, histrelin acetate, ibritumomab, interferon alfa-2a, interferon alfa-2b, lenalidomide, levamisole, mesna, methoxsalen, nandrolone, nelarabine, nofetumomab, oprelvekin, palifermin, pamidronate, pegademase, pegaspargase, pegfilgrastim, pemetrexed disodium, plicamycin, porfimer sodium, quinacrine, rasburicase, sargramostim, temozolomide, VM-26, 6-TG, toremifene, tretinoin, ATRA, valrubicin, zoledronate, and zoledronic acid, and pharmaceutically acceptable salts thereof.

[0106] Chemotherapeutic agents also include hydrocortisone, hydrocortisone acetate, cortisone acetate, tixocortol pivalate, triamcinolone acetonide, triamcinolone alcohol, mometasone, amcinonide, budesonide, desonide, fluocinonide, fluocinolone acetonide, betamethasone, betamethasone sodium phosphate, dexamethasone, dexamethasone sodium phosphate, fluocortolone, hydrocortisone-17-butyrate, hydrocortisone-17-valerate, aclometasone dipropionate, betamethasone valerate, betamethasone dipropionate, prednicarbate, clobetasone-17-butyrate, clobetasol-17-propionate, fluocortolone caproate, fluocortolone pivalate and fluprednidene acetate; immune selective anti-inflammatory peptides (ImSAIDs) such as phenylalanine-glutamine-glycine (FEG) and its D-isomeric form (feG) (IMULAN BioTherapeutics, LLC); anti-rheumatic drugs such as azathioprine, ciclosporin (cyclosporine A), D-penicillamine, gold salts, hydroxychloroquine, leflunomideminocycline, sulfasalazine, tumor necrosis factor alpha (TNFα) blockers such as etanercept (Enbrel), infliximab (Remicade), adalimumab (Humira), certolizumab pegol (Cimzia), golimumab (Simponi), Interleukin 1 (IL-1) blockers such as anakinra (Kineret), T

cell costimulation blockers such as abatacept (Orencia), Interleukin 6 (IL-6) blockers such as tocilizumab (ACTEMERA®); Interleukin 13 (IL-13) blockers such as lebrikizumab; Interferon alpha (IFN) blockers such as Rontalizumab; Beta 7 integrin blockers such as rhuMAb Beta7; IgE pathway blockers such as Anti-M1 prime; Secreted homotrimeric LTa3 and membrane bound heterotrimer LTa1/β2 blockers such as Anti-lymphotoxin alpha (LTa); radioactive isotopes (e.g., At211, I131, I125, Y90, Re186, Re188, Sm153, Bi212, P32, Pb212 and radioactive isotopes of Lu); miscellaneous investigational agents such as thioplatin, PS-341, phenylbutyrate, ET-18- OCH3, or farnesyl transferase inhibitors (L-739749, L-744832); polyphenols such as quercetin, resveratrol, piceatannol, epigallocatechine gallate, theaflavins, flavanols, procyanidins, betulinic acid and derivatives thereof; autophagy inhibitors such as chloroquine; delta-9-tetrahydrocannabinol (dronabinol, MARINOL®); beta-lapachone; lapachol; colchicines; betulinic acid; acetylcamptothecin, scopolectin, and 9-aminocamptothecin); podophyllotoxin; tegafur (UFTORAL®); bexarotene (TARGRETIN®); bisphosphonates such as clodronate (for example, BONEFOS® or OSTAC®), etidronate (DIDROCAL®), NE-58095, zoledronic acid/zoledronate (ZOMETA®), alendronate (FOSAMAX®), pamidronate (AREDIA®), tiludronate (SKELID®), or risedronate (ACTONEL®); and epidermal growth factor receptor (EGF-R); vaccines such as THERATOPE® vaccine; perifosine, COX-2 inhibitor (e.g. celecoxib or etoricoxib), proteosome inhibitor (e.g. PS341); CCI-779; tipifarnib (R11577); orafenib, ABT510; Bcl-2 inhibitor such as oblimersen sodium (GENASENSE®); pixantrone; farnesyltransferase inhibitors such as lonafarnib (SCH 6636, SARASARTM); and pharmaceutically acceptable salts, acids or derivatives of any of the above; as well as combinations of two or more of the above such as CHOP, an abbreviation for a combined therapy of cyclophosphamide, doxorubicin, vincristine, and prednisolone; and FOLFOX, an abbreviation for a treatment regimen with oxaliplatin (ELOXATINTM) combined with 5-FU and leucovorin.

[0107] Chemotherapeutic agents also include non-steroidal anti-inflammatory drugswith analgesic, antipyretic and anti-inflammatory effects. NSAIDs include non-selective inhibitors of the enzyme cyclooxygenase. Specific examples of NSAIDs include aspirin, propionic acid derivatives such as ibuprofen, fenoprofen, ketoprofen, flurbiprofen, oxaprozin and naproxen, acetic acid derivatives such as indomethacin, sulindac, etodolac, diclofenac, enolic acid derivatives such as piroxicam, meloxicam, tenoxicam, droxicam, lornoxicam and isoxicam, fenamic acid derivatives such as mefenamic acid, meclofenamic acid, flufenamic acid, tolfenamic acid, and COX-2 inhibitors such as celecoxib, etoricoxib, lumiracoxib, parecoxib,

rofecoxib, rofecoxib, and valdecoxib. NSAIDs can be indicated for the symptomatic relief of conditions such as rheumatoid arthritis, osteoarthritis, inflammatory arthropathies, ankylosing spondylitis, psoriatic arthritis, Reiter's syndrome, acute gout, dysmenorrhoea, metastatic bone pain, headache and migraine, postoperative pain, mild-to-moderate pain due to inflammation and tissue injury, pyrexia, ileus, and renal colic.

[0108] The term "cytokine" is a generic term for proteins released by one cell population that act on another cell as intercellular mediators. Examples of such cytokines are lymphokines, monokines; interleukins (ILs) such as IL-1, IL-1a, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-11, IL-12, IL-15; a tumor necrosis factor such as TNF-α or TNF-β; and other polypeptide factors including LIF and kit ligand (KL) and gamma interferon. As used herein, the term cytokine includes proteins from natural sources or from recombinant cell culture and biologically active equivalents of the native-sequence cytokines, including synthetically produced small-molecule entities and pharmaceutically acceptable derivatives and salts thereof.

[0109] The term "PD-1 axis binding antagonist" is a molecule that inhibits the interaction of a PD-1 axis binding partner with either one or more of its binding partner, so as to remove T-cell dysfunction resulting from signaling on the PD-1 signaling axis - with a result being to restore or enhance T-cell function (e.g., proliferation, cytokine production, target cell killing). As used herein, a PD-1 axis binding antagonist includes a PD-1 binding antagonist, a PD-L1 binding antagonist and a PD-L2 binding antagonist.

[0110] The term "PD-1 binding antagonists" is a molecule that decreases, blocks, inhibits, abrogates or interferes with signal transduction resulting from the interaction of PD-1 with one or more of its binding partners, such as PD-L1, PD-L2. In some embodiments, the PD-1 binding antagonist is a molecule that inhibits the binding of PD-1 to its binding partners. In a specific aspect, the PD-1 binding antagonist inhibits the binding of PD-1 to PD-L1 and/or PD-L2. For example, PD-1 binding antagonists include anti-PD-1 antibodies, antigen binding fragments thereof, immunoadhesins, fusion proteins, oligopeptides and other molecules that decrease, block, inhibit, abrogate or interfere with signal transduction resulting from the interaction of PD-1 with PD-L1 and/or PD-L2. In one embodiment, a PD-1 binding antagonist reduces the negative co-stimulatory signal mediated by or through cell surface proteins expressed on T lymphocytes mediated signaling through PD-1 so as render a dysfunctional T-cell less dysfunctional (e.g. , enhancing effector responses to antigen recognition). In some embodiments, the PD-1 binding antagonist is an anti-PD-1 antibody. In a specific aspect, a PD-1 binding antagonist is MDX- 1 106 described herein. In another

specific aspect, a PD-1 binding antagonist is Merck 3745 described herein. In another specific aspect, a PD-1 binding antagonist is CT-011 described herein.

[0111] The term "PD-L1 binding antagonists" is a molecule that decreases, blocks, inhibits, abrogates or interferes with signal transduction resulting from the interaction of PD-L1 with either one or more of its binding partners, such as PD-1, B7-1. In some embodiments, a PD-L1 binding antagonist is a molecule that inhibits the binding of PD-L1 to its binding partners. In a specific aspect, the PD-L1 binding antagonist inhibits binding of PD-L1 to PD-1 and/or B7-1. In some embodiments, the PD-L1 binding antagonists include anti-PD-L1 antibodies, antigen binding fragments thereof, immunoadhesins, fusion proteins, oligopeptides and other molecules that decrease, block, inhibit, abrogate or interfere with signal transduction resulting from the interaction of PD-L1 with one or more of its binding partners, such as PD-1, B7- 1. In one embodiment, a PD-L1 binding antagonist reduces the negative co-stimulatory signal mediated by or through cell surface proteins expressed on T lymphocytes mediated signaling through PD-L1 so as to render a dysfunctional T-cell less dysfunctional (e.g. , enhancing effector responses to antigen recognition). In some embodiments, a PD-L1 binding antagonist is an anti-PD-L1 antibody. In a specific aspect, an anti-PD-L1 antibody is YW243.55.S70 described herein. In another specific aspect, an anti-PD-L1 antibody is MDX- 1 105 described herein. In still another specific aspect, an anti-PD-L1 antibody is MPDL3280A described herein.

[0112] The term "PD-L2 binding antagonists" is a molecule that decreases, blocks, inhibits, abrogates or interferes with signal transduction resulting from the interaction of PD-L2 with either one or more of its binding partners, such as PD-1. In some embodiments, a PD-L2 binding antagonist is a molecule that inhibits the binding of PD-L2 to its binding partners. In a specific aspect, the PD-L2 binding antagonist inhibits binding of PD-L2 to PD-1. In some embodiments, the PD-L2 antagonists include anti-PD-L2 antibodies, antigen binding fragments thereof, immunoadhesins, fusion proteins, oligopeptides and other molecules that decrease, block, inhibit, abrogate or interfere with signal transduction resulting from the interaction of PD-L2 with either one or more of its binding partners, such as PD-1. In one embodiment, a PD-L2 binding antagonist reduces the negative co-stimulatory signal mediated by or through cell surface proteins expressed on T lymphocytes mediated signaling through PD-L2 so as render a dysfunctional T-cell less dysfunctional (e.g., enhancing effector responses to antigen recognition). In some embodiments, a PD-L2 binding antagonist is an immunoadhesin.

[0113] The term "phagocytosis" means the internalization of cells or particulate matter by cells. In some embodiments, the phagocytic cells or phagocytes are macrophages or neutrophils. In some embodiments, the cells are cells that express human OX40. Methods for assaying phagocytosis are known in the art and include use of microscopy to detect the presence of cells internalized within another cells. In other embodiments, phagocytosis is detected using FACS, e.g., by detecting presence of a detectably labeled cell within another cell (which may be detectably labeled, e.g., with a different label than the first cell).

[0114] The phrase "does not possess substantial activity" or "substantially no activity" with respect to an antibody, as used herein, means the antibody does not exhibit an activity that is above background level (in some embodiments, that is above background level that is statistically significant). The phrase "little to no activity" with respect to an antibody, as used herein, means the antibody does not display a biologically meaningful amount of a function. The function can be measured or detected according to any assay or technique known in the art, including, e.g., those described herein. In some embodiments, antibody function is stimulation of effector T cell proliferation and/or cytokine secretion.

[0115] The term "biomarker" or "marker" as used herein refers generally to a molecule, including a gene, mRNA, protein, carbohydrate structure, or glycolipid, the expression of which in or on a tissue or cell or secreted can be detected by known methods (or methods disclosed herein) and is predictive or can be used to predict (or aid prediction) for a cell, tissue, or patient's responsiveness to treatment regimes. In some embodiments, a biomarker may refer to a gene or protein, e.g., the level of expression of the gene or protein detected in one or more cells. In some embodiments, a biomarker may refer to a cell type of interest, e.g., the number of a cell type of interest detected in one or more samples.

[0116] By "patient sample" is meant a collection of cells or fluids obtained from a cancer patient. The source of the tissue or cell sample may be solid tissue as from a fresh, frozen and/or preserved organ or tissue sample or biopsy or aspirate; blood or any blood constituents; bodily fluids such as cerebrospinal fluid, amniotic fluid, peritoneal fluid, or interstitial fluid; cells from any time in gestation or development of the subject. The tissue sample may contain compounds which are not naturally intermixed with the tissue in nature such as preservatives, anticoagulants, buffers, fixatives, nutrients, antibiotics, or the like. Examples of tumor samples herein include, but are not limited to, tumor biopsy, fine needle aspirate, bronchiolar lavage, pleural fluid, sputum, urine, a surgical specimen, circulating tumor cells, serum, plasma, circulating plasma proteins, ascitic fluid, primary cell cultures or

cell lines derived from tumors or exhibiting tumor-like properties, as well as preserved tumor samples, such as formalin-fixed, paraffin-embedded tumor samples or frozen tumor samples.

[0117] The phrase "based on expression of" when used herein means that information about expression level or presence or absence of expression (e.g., presence or absence or prevalence of (e.g., percentage of cells displaying) of the one or more biomarkers herein (e.g., presence or absence of or amount or prevalence of FcR-expressing cells, or e.g., presence or absence or amount or prevalence of human effector cells) is used to inform a treatment decision, information provided on a package insert, or marketing/promotional guidance etc.

[0118] A cancer or biological sample which "has human effector cells" is one which, in a diagnostic test, has human effector cells present in the sample (e.g., infiltrating human effector cells).

[0119] A cancer or biological sample which "has FcR-expressing cells" is one which, in a diagnostic test, has FcR-expressing present in the sample (e.g., infiltrating FcR-expressing cells). In some embodiments, FcR is FcγR. In some embodiments, FcR is an activating FcγR.

[0120] The phrase "recommending a treatment" as used herein refers to using the information or data generated relating to the level or presence of c-met in a sample of a patient to identify the patient as suitably treated or not suitably treated with a therapy. In some embodiments the therapy may comprise c-met antibody (e.g., onartuzumab). In some embodiments, the therapy may comprise VEGF antagonist (e.g., bevacizumab). In some embodiments, the therapy may comprise anti-human OX40 agonist antibody. The information or data may be in any form, written, oral or electronic. In some embodiments, using the information or data generated includes communicating, presenting, reporting, storing, sending, transferring, supplying, transmitting, delivering, dispensing, or combinations thereof. In some embodiments, communicating, presenting, reporting, storing, sending, transferring, supplying, transmitting, delivering, dispensing, or combinations thereof are performed by a computing device, analyzer unit or combination thereof. In some further embodiments, communicating, presenting, reporting, storing, sending, transferring, supplying, transmitting, dispensing, or combinations thereof are performed by an individual (e.g., a laboratory or medical professional). In some embodiments, the information or data includes a comparison of the amount or prevalence of FcR expressing cells to a reference level. In some embodiments, the information or data includes a comparison of the amount or prevalence of human effector cells to a reference level. In some embodiments, the information or data includes an indication that human effector cells or FcR-expressing cells are present or absent in the sample. In some embodiments, the information or data includes

an indication that FcR-expressing cells and/or human effector cells are present in a particular percentage of cells (e.g., high prevalence). In some embodiments, the information or data includes an indication that the patient is suitably treated or not suitably treated with a therapy comprising anti-human OX40 agonist antibody.

[0121] The term "*detection*" includes any means of detecting, including direct and indirect detection.

[0122] The "amount" or "level" of a biomarker associated with an increased clinical benefit to an individual is a detectable level in a biological sample. These can be measured by methods known to one skilled in the art and also disclosed herein. The expression level or amount of biomarker assessed can be used to determine the response to the treatment.

[0123] "Elevated expression," "elevated expression levels," or "elevated levels" refers to an increased expression or increased levels of a biomarker in an individual relative to a control, such as an individual or individuals who are not suffering from the disease or disorder (e.g., cancer), a tumor with a known responsiveness to a treatment (e.g., with an OX40 agonist), an internal control (e.g., housekeeping biomarker), or a reference number (e.g., a set threshold amount, such as a threshold based on clinical outcome data).

[0124] "Reduced expression," "reduced expression levels," or "reduced levels" refers to a decrease expression or decreased levels of a biomarker in an individual relative to a control, such as an individual or individuals who are not suffering from the disease or disorder (e.g., cancer), a tumor with a known responsiveness to a treatment (e.g., with an OX40 agonist), an internal control (e.g., housekeeping biomarker), or a reference number (e.g., a set threshold amount, such as a threshold based on clinical outcome data). In some embodiments, reduced expression is little or no expression.

[0125] The terms "*housekeeping gene*" or "*housekeeping biomarker*" as used herein may refer to any gene or genes thought to be constitutively expressed in a cell in normal and/or pathological states. Such a gene may be used, for example, as a reference, since its expression is detectable at a consistent amount across different physiological conditions. In some embodiments, a housekeeping gene may encode a protein required for basic cellular function and/or maintenance.

[0126] The term "diagnosis" is used herein to refer to the identification or classification of a molecular or pathological state, disease or condition (e.g., cancer). For example, "diagnosis" may refer to identification of a particular type of cancer. "Diagnosis" may also refer to the classification of a particular subtype of cancer, e.g., by histopathological criteria,

or by molecular features (e.g., a subtype characterized by expression of one or a combination of biomarkers (e.g., particular genes or proteins encoded by said genes)).

[0127] The term "aiding diagnosis" is used herein to refer to methods that assist in making a clinical determination regarding the presence, or nature, of a particular type of symptom or condition of a disease or disorder (e.g., cancer). For example, a method of aiding diagnosis of a disease or condition (e.g., cancer) can comprise measuring certain biomarkers in a biological sample from an individual.

[0128] The term "sample," as used herein, refers to a composition that is obtained or derived from a subject and/or individual of interest that contains a cellular and/or other molecular entity that is to be characterized and/or identified, for example based on physical, biochemical, chemical and/or physiological characteristics. For example, the phrase "disease sample" and variations thereof refers to any sample obtained from a subject of interest that would be expected or is known to contain the cellular and/or molecular entity that is to be characterized. Samples include, but are not limited to, primary or cultured cells or cell lines, cell supernatants, cell lysates, platelets, serum, plasma, vitreous fluid, lymph fluid, synovial fluid, follicular fluid, seminal fluid, amniotic fluid, milk, whole blood, blood-derived cells, urine, cerebrospinal fluid, saliva, sputum, tears, perspiration, mucus, tumor lysates, and tissue culture medium, tissue extracts such as homogenized tissue, tumor tissue, cellular extracts, and combinations thereof.

[0129] By "tissue sample" or "cell sample" is meant a collection of similar cells obtained from a tissue of a subject or individual. The source of the tissue or cell sample may be solid tissue as from a fresh, frozen and/or preserved organ, tissue sample, biopsy, and/or aspirate; blood or any blood constituents such as plasma; bodily fluids such as cerebral spinal fluid, amniotic fluid, peritoneal fluid, or interstitial fluid; cells from any time in gestation or development of the subject. The tissue sample may also be primary or cultured cells or cell lines. Optionally, the tissue or cell sample is obtained from a disease tissue/organ. The tissue sample may contain compounds which are not naturally intermixed with the tissue in nature such as preservatives, anticoagulants, buffers, fixatives, nutrients, antibiotics, or the like.

[0130] As used herein, a "section" of a tissue sample is meant a single part or piece of a tissue sample, *e.g.* a thin slice of tissue or cells cut from a tissue sample. It is understood that multiple sections of tissue samples may be taken and subjected to analysis according to the present invention, provided that it is understood that the present invention comprises a method whereby the same section of tissue sample is analyzed at both morphological and molecular levels, or is analyzed with respect to protein or nucleic acid.

[0131] By “correlate” or “correlating” is meant comparing, in any way, the performance and/or results of a first analysis or protocol with the performance and/or results of a second analysis or protocol. For example, one may use the results of a first analysis or protocol in carrying out a second protocols and/or one may use the results of a first analysis or protocol to determine whether a second analysis or protocol should be performed. With respect to the embodiment of polypeptide analysis or protocol, one may use the results of the polypeptide expression analysis or protocol to determine whether a specific therapeutic regimen should be performed. With respect to the embodiment of polynucleotide analysis or protocol, one may use the results of the polynucleotide expression analysis or protocol to determine whether a specific therapeutic regimen should be performed.

[0132] “Individual response” or “response” can be assessed using any endpoint indicating a benefit to the individual, including, without limitation, (1) inhibition, to some extent, of disease progression (e.g., cancer progression), including slowing down and complete arrest; (2) a reduction in tumor size; (3) inhibition (i.e., reduction, slowing down or complete stopping) of cancer cell infiltration into adjacent peripheral organs and/or tissues; (4) inhibition (i.e. reduction, slowing down or complete stopping) of metastasis; (5) relief, to some extent, of one or more symptoms associated with the disease or disorder (e.g., cancer); (6) increase or extend in the length of survival, including overall survival and progression free survival; and/or (9) decreased mortality at a given point of time following treatment.

[0133] An “effective response” of a patient or a patient's “responsiveness” to treatment with a medicament and similar wording refers to the clinical or therapeutic benefit imparted to a patient at risk for, or suffering from, a disease or disorder, such as cancer. In one embodiment, such benefit includes any one or more of: extending survival (including overall survival and progression free survival); resulting in an objective response (including a complete response or a partial response); or improving signs or symptoms of cancer.

[0134] By “extending survival” is meant increasing overall or progression free survival in a treated patient relative to an untreated patient (i.e. relative to a patient not treated with the medicament), or relative to a patient who does not express a biomarker at the designated level, and/or relative to a patient treated with an approved anti-tumor agent. An objective response refers to a measurable response, including complete response (CR) or partial response (PR).

III. OX40 Agonists

[0135] Provided herein are methods for predicting responsiveness of a subject having cancer to an OX40 agonist treatment. These methods are based in part on the discovery described herein that the expression level of specific biomarkers correlates with responsiveness to OX40 agonist treatment. For example, an increased expression level of genes such as CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R correlates with responsiveness to OX40 agonist treatment. Additionally, a decreased expression level of genes such as CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1 was also found to correlate with responsiveness to OX40 agonist treatment.

[0136] Further provided herein are methods for monitoring pharmacodynamic activity of an OX40 agonist treatment by measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the subject has been treated with an OX40 agonist, and where the one or more marker genes are selected from ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1, PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and TNFSF4; and determining the treatment as demonstrating pharmacodynamic activity based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates pharmacodynamic activity to the OX40 agonist treatment. These methods are based in part on the discovery described herein that the expression level of specific biomarkers is upregulated upon OX40 agonist treatment.

[0137] Yet further provided herein are methods monitoring responsiveness of a subject to an OX40 agonist treatment by measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the subject has been treated with an OX40 agonist, and where the one or more marker genes are selected from BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, H2-aa, H2-d, H2-k, ICOS, IL10, PDCA1, and TNFRSF18; and classifying the subject as responsive or non-responsive to said treatment based on the expression level of said one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates a responsive subject. These methods are based in part on the discovery

described herein that the expression level of specific biomarkers is upregulated upon OX40 agonist treatment specifically in tumors responsive to such treatment.

[0138] Any OX40 agonists known in the art may be used in the methods described herein. In one aspect, an OX40 agonist is an isolated antibody that binds to human OX40. Antibodies suitable for use in the methods of the invention include antibodies that bind to human OX40. Descriptions of anti-OX40 antibodies (*e.g.,* anti-human OX40 agonist antibodies) may be found in US PG Pub. No. US2015/0307617 and International Publication No. WO/2015/153513, which are each incorporated by reference herein in their entirety.

[0139] In some embodiments, the anti-human OX40 agonist antibody binds human OX40 with an affinity of less than or equal to about 0.45 nM. In some embodiments, the anti-human OX40 antibody binds human OX40 with an affinity of less than or equal to about 0.4 nM. In some embodiments, the anti-human OX40 antibody binds human OX40 with an affinity of less than or equal to about 0.5nM. In some embodiments, the binding affinity is determined using radioimmunoassay.

[0140] In some embodiments, the anti-human OX40 agonist antibody binds human OX40 and cynomolgus OX40. In some embodiments, binding is determined using a FACS assay. In some embodiments, binding to human OX40 has an EC50 of about 0.2 ug/ml. In some embodiments, binding to human OX40 has an EC50 of about 0.3 ug/ml or lower. In some embodiments, binding to cynomolgus OX40 has an EC50 of about 1.5 ug/ml. In some embodiments, binding to cynomolgus OX40 has an EC50 of about 1.4 ug/ml.

[0141] In some embodiments, the anti-human OX40 agonist antibody does not bind to rat OX40 or mouse OX40.

[0142] In some embodiments, the anti-human OX40 agonist antibody is a depleting anti-human OX40 antibody (e.g., depletes cells that express human OX40). In some embodiments, the human OX40 expressing cells are CD4+ effector T cells. In some embodiments, the human OX40 expressing cells are Treg cells. In some embodiments, depleting is by ADCC and/or phagocytosis. In some embodiments, the antibody mediates ADCC by binding FcγR expressed by a human effector cell and activating the human effector cell function. In some embodiments, the antibody mediates phagocytosis by binding FcγR expressed by a human effector cell and activating the human effector cell function. Exemplary human effector cells include, e.g., macrophage, natural killer (NK) cells, monocytes, neutrophils. In some embodiments, the human effector cell is macrophage. In some embodiments, the human effector cell is NK cells. In some embodiments, depletion is not by apoptosis.

[0143] In some embodiments, the anti-human OX40 agonist antibody has a functional Fc region. In some embodiments, effector function of a functional Fc region is ADCC. In some embodiments, effector function of a functional Fc region is phagocytosis. In some embodiments, effector function of a functional Fc region is ADCC and phagocytosis. In some embodiments, the Fc region is human IgG1. In some embodiments, the Fc region is human IgG4.

[0144] In some embodiments, the anti-human OX40 agonist antibody does not induce apoptosis in OX40-expressing cells (e.g., Treg). In some embodiments, apoptosis is assayed using an antibody concentration of 30ug/ml, e.g., by determining whether apoptosis has occurred using annexin V and proprodium iodide stained Treg.

[0145] In some embodiments, the anti-human OX40 agonist antibody enhances CD4+ effector T cell function, for example, by increasing CD4+ effector T cell proliferation and/or increasing gamma interferon production by the CD4+ effector T cell (for example, as compared to proliferation and/or cytokine production prior to treatment with anti-human OX40 agonist antibody). In some embodiments, the cytokine is gamma interferon. In some embodiments, the anti-human OX40 agonist antibody increases number of intratumoral (infiltrating) CD4+ effector T cells (e.g., total number of CD4+ effector T cells, or e.g., percentage of CD4+ cells in CD45+ cells), e.g., as compared to number of intratumoral (infiltrating) CD4+ T cells prior to treatment with anti-human OX40 agonist antibody. In some embodiments, the anti-human OX40 agonist antibody increases number of intratumoral (infiltrating) CD4+ effector T cells that express gamma interferon (e.g., total gamma interferon expressing CD4+ cells, or e.g., percentage of gamma interferon expressing CD4+ cells in total CD4+ cells), e.g., as compared to number of intratumoral (infiltrating) CD4+ T cells that express gamma interferon prior to treatment with anti-human OX40 agonist antibody.

[0146] In some embodiments, the anti-human OX40 agonist antibody increases number of intratumoral (infiltrating) CD8+ effector T cells (e.g., total number of CD8+ effector T cells, or e.g., percentage of CD8+ in CD45+ cells), e.g., as compared to number of intratumoral (infiltrating) CD8+ T effector cells prior to treatment with anti-human OX40 agonist antibody. In some embodiments, the anti-human OX40 agonist antibody increases number of intratumoral (infiltrating) CD8+ effector T cells that express gamma interferon (e.g., percentage of CD8+ cells that express gamma interferon in total CD8+ cells), e.g., compared to number of intratumoral (infiltrating) CD8+ T cells that express gamma interferon prior to treatment with anti-human OX40 agonist antibody.

[0147] In some embodiments, the anti-human OX40 agonist antibody enhances memory T cell function, for example by increasing memory T cell proliferation and/or increasing cytokine production by the memory cell. In some embodiments, the cytokine is gamma interferon.

[0148] In some embodiments, the anti-human OX40 agonist antibody inhibits Treg function, for example, by decreasing Treg suppression of effector T cell function (e.g., effector T cell proliferation and/or effector T cell cytokine secretion). In some embodiments, the effector T cell is a CD4+ effector T cell. In some embodiments, the anti-human OX40 agonist antibody reduces the number of intratumoral (infiltrating) Treg (e.g., total number of Treg or e.g., percentage of Fox3p+ cells in CD4+ cells).

[0149] In some embodiments, the anti-human OX40 agonist antibody is engineered to increase effector function (e.g., compared to effector function in a wild-type IgG1). In some embodiments, the antibody has increased binding to a Fcγ receptor. In some embodiments, the antibody lacks fucose attached (directly or indirectly) to the Fc region. For example, the amount of fucose in such antibody may be from 1% to 80%, from 1% to 65%, from 5% to 65% or from 20% to 40%. In some embodiments, the Fc region comprises bisected oligosaccharides, e.g., in which a biantennary oligosaccharide attached to the Fc region of the antibody is bisected by GlcNAc. In some embodiments, the antibody comprises an Fc region with one or more amino acid substitutions which improve ADCC, e.g., substitutions at positions 298, 333, and/or 334 of the Fc region (EU numbering of residues).

[0150] In some embodiments, the anti-human OX40 agonist antibody increases OX40 signal transduction in a target cell that expresses OX40. In some embodiments, OX40 signal transduction is detected by monitoring NFkB downstream signaling.

[0151] In some embodiments, the anti-human OX40 agonist antibody is stable after treatment at 40C for two weeks.

[0152] In some embodiments, the anti-human OX40 agonist antibody binds human effector cells, e.g., binds FcγR (e.g., an activating FcγR) expressed by human effector cells. In some embodiments, the human effector cell performs (is capable of performing) ADCC effector function. In some embodiments, the human effector cell performs (is capable of performing) phagocytosis effector function.

[0153] In some embodiments, the anti-human OX40 agonist antibody comprising a variant IgG1 Fc polypeptide comprising a mutation that eliminates binding to human effector cells (e.g., a DANA mutation) has diminished activity (e.g., CD4+ effector T cell function, e.g., proliferation), relative to anti-human OX40 agonist antibody comprising native sequence

IgG1 Fc portion. In some embodiment, the anti-human OX40 agonist antibody comprising a variant IgG1 Fc polypeptide comprising a mutation that eliminates binding to human effector cells (e.g., a DANA mutation) does not possess substantial activity (e.g., CD4+ effector T cell function, e.g., proliferation).

[0154] In some embodiments, antibody cross-linking is required for anti-human OX40 agonist antibody function. In some embodiments, function is stimulation of CD4+ effector T cell proliferation. In some embodiments, antibody cross-linking is determined by providing anti-human OX40 agonist antibody adhered on a solid surface (e.g., a cell culture plate). In some embodiments, antibody cross-linking is determined by introducing a mutation in the antibody's IgG1 Fc portion (e.g., a DANA mutation) and testing function of the mutant antibody.

[0155] In some embodiments, the anti-human OX40 agonist antibody competes for binding to human OX40 with OX40L. In some embodiments, addition of OX40L does not enhance anti-human OX40 antibody function in an in vitro assay.

[0156] According to another embodiment, the anti-human OX40 agonist antibodies include any one, any combination, or all of the following properties: (1) binds human OX40 with an affinity of less than or equal to about 0.45 nM, in some embodiments, binds human OX40 with an affinity of less than or equal to about 0.4 nM, in some embodiments, binds human OX40 with an affinity of less than or equal to about 0.5nM, in some embodiments, the binding affinity is determined using radioimmunoassay; (2) binds human OX40 and cynomolgus OX40, in some embodiments, binding is determined using a FACS assay, (3) binds human OX40 with an EC50 of about 0.2 ug/ml, in some embodiments, binds to human OX40 has an EC50 of about 0.3 ug/ml or lower, in some embodiments, binds to cynomolgus OX40 with an EC50 of about 1.5 ug/ml, in some embodiments, binds to cynomolgus OX40 has an EC50 of about 1.4 ug/ml, (4) does not substantially bind to rat OX40 or mouse OX40, (6) is a depleting anti-human OX40 antibody (e.g., depletes cells that express human OX40), in some embodiments, the cells are CD4+ effector T cells and/or Treg cells, (7) enhances CD4+ effector T cell function, for example, by increasing CD4+ effector T cell proliferation and/or increasing gamma interferon production by the CD4+ effector T cell (for example, as compared to proliferation and/or cytokine production prior to treatment with anti-human OX40 agonist antibody), (8) enhances memory T cell function, for example by increasing memory T cell proliferation and/or increasing cytokine production by the memory cell, (9) inhibits Treg function, for example, by decreasing Treg suppression of effector T cell function (e.g., effector T cell proliferation and/or effector T cell cytokine secretion). In some

embodiments, the effector T cell is a CD4+ effector T cell, (10) increases OX40 signal transduction in a target cell that expresses OX40 (in some embodiments, OX40 signal transduction is detected by monitoring NFkB downstream signaling), (11) is stable after treatment at 40C for two weeks, (12) binds human effector cells, e.g., binds FcγR expressed by human effector cells, (13) anti-human OX40 agonist antibody comprising a variant IgG1 Fc polypeptide comprising a mutation that eliminates binding to human effector cells (e.g., N297G) has diminished activity (e.g., CD4+ effector T cell function, e.g., proliferation), relative to anti-human OX40 agonist antibody comprising native sequence IgG1 Fc portion, in some embodiment, the anti-human OX40 agonist antibody comprising a variant IgG1 Fc polypeptide comprising a mutation that eliminates binding to human effector cells (e.g., N297G) does not possess substantial activity (e.g., CD4+ effector T cell function, e.g., proliferation), (14) antibody cross-linking (e.g., by Fc receptor binding) is required for anti-human OX40 agonist antibody function.

[0157] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7.

[0158] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4. In one embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:7. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, HVR-L3 comprising the amino acid sequence of SEQ ID NO:7, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:3. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4.

[0159] In another aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7. In one embodiment, the antibody comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7.

[0160] In another aspect, an anti-human OX40 agonist antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:4; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7.

[0161] In another aspect, the invention provides an anti-human OX40 agonist antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:7.

[0162] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:26.

[0163] In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:26. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, HVR-L3 comprising the amino acid sequence of SEQ ID NO:26, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:3.

[0164] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:4; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:26.

[0165] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:26.

[0166] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:27.

[0167] In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:27. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, HVR-L3 comprising the amino acid sequence of SEQ ID NO:27, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:3.

[0168] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:4; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:27.

[0169] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:27.

[0170] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, 10, 11, 12, 13 or 14; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, 15, or 19; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7, 22, 23, 24, 25, 26, 27, or 28.

[0171] In one aspect, the invention provides an antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO: 2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 3, 10, 11, 12, 13 or 14; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO: 4, 15, or 19. In one embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO: 4, 15, or 19. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:4, 15, or 19 and HVR-L3 comprising the amino acid sequence of SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO: 4, 15, or 19, HVR-L3 comprising the amino acid sequence of SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28, and HVR-H2 comprising the amino acid sequence of SEQ ID NO: 3, 10, 11, 12, 13 or 14. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO: 2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 3, 10, 11, 12, 13 or 14; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO: 4, 15, or 19.

[0172] In another aspect, the invention provides an antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO: 5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28. In one embodiment, the antibody comprises (a) HVR-L1 comprising the

amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28.

[0173] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO: 2, 8 or 9, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 3, 10, 11, 12, 13 or 14, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO: 4, 15, or 19; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28.

[0174] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO: 2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 3, 10, 11, 12, 13 or 14; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO: 4, 15, or 19; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO: 7, 22, 23, 24, 25, 26, 27, or 28.

[0175] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:172; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:173; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:174; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:175. In some embodiment, HVR-H2 is not DMYPDAAAASYNQKFRE (SEQ ID NO:222).In some embodiments, HVR-H3 is not APRWAAAA (SEQ ID NO:223). In some embodiments, HVR-L3 is not QAAAAAAAT (SEQ ID NO:224).

[0176] In one aspect, the invention provides an antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:172; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:173; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:174. In one embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ

ID NO:174. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:174 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:175. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:174, HVR-L3 comprising the amino acid sequence of SEQ ID NO:175, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:173. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:172; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:173; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:174. In some embodiment, HVR-H2 is not DMYPDAAAASYNQKFRE (SEQ ID NO:222).In some embodiments, HVR-H3 is not APRWAAAA (SEQ ID NO:223). In some embodiments, HVR-L3 is not QAAAAAAAT (SEQ ID NO:224).

[0177] In another aspect, the invention provides an antibody comprising (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:175. In some embodiments, HVR-L3 is not QAAAAAAAT (SEQ ID NO:224).

[0178] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:172, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:173, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:174; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:175.

[0179] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:172; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:173; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:174; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:175. In some embodiment, HVR-H2 is not DMYPDAAAASYNQKFRE (SEQ ID NO:222).In some embodiments, HVR-H3 is not APRWAAAA (SEQ ID NO:223). In some embodiments, HVR-L3 is not QAAAAAAAT (SEQ ID NO:224).

[0180] All possible combinations of the above substitutions are encompassed by the consensus sequences of SEQ ID NO:172, 173, 174 and 175.

[0181] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0182] In one aspect, the invention provides an antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33. In one embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:42. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33, HVR-L3 comprising the amino acid sequence of SEQ ID NO:42, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:30. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33.

[0183] In another aspect, the invention provides an antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42. In one embodiment, the antibody comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0184] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:33; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid

sequence of SEQ ID NO:37, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0185] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:42.

[0186] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:40; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0187] In another aspect, the invention provides an antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:40; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42. In one embodiment, the antibody comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:40; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0188] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:33; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:40, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0189] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2

comprising the amino acid sequence of SEQ ID NO:40; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:42.

[0190] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30, 31, or 32; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39, 40 or 41; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42, 43, or 44.

[0191] In one aspect, the invention provides an antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 30, 31, or 32; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33 and HVR-L3 comprising the amino acid sequence of SEQ ID NO: 42, 43, or 44. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33, HVR-L3 comprising the amino acid sequence of SEQ ID NO: 42, 43, or 44, and HVR-H2 comprising the amino acid sequence of SEQ ID NO: 39, 40 or 41. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30, 31, or 32; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33.

[0192] In another aspect, the invention provides an antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO: 39, 40 or 41; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 42, 43, or 44. In one embodiment, the antibody comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO: 39, 40 or 41; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 42, 43, or 44.

[0193] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 30, 31, or 32, and (iii) HVR-H3 comprising an amino

acid sequence selected from SEQ ID NO:33; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO: 39, 40 or 41, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO: 42, 43, or 44.

[0194] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO: 30, 31, or 32; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO: 39, 40 or 41; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO: 42, 43, or 44.

[0195] In one aspect, the invention provides an anti-human OX40 agonist antibody comprising at least one, two, three, four, five, or six HVRs selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:175; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:177; and (f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:178.

[0196] In one aspect, the invention provides an antibody comprising at least one, at least two, or all three VH HVR sequences selected from (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:175; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33. In another embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33 and HVR-L3 comprising the amino acid sequence of SEQ ID NO:177. In a further embodiment, the antibody comprises HVR-H3 comprising the amino acid sequence of SEQ ID NO:33, HVR-L3 comprising the amino acid sequence of SEQ ID NO:178, and HVR-H2 comprising the amino acid sequence of SEQ ID NO:176. In a further embodiment, the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:176; and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33.

[0197] In another aspect, the invention provides an antibody comprising at least one, at least two, or all three VL HVR sequences selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:177; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:177. In

one embodiment, the antibody comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:177; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:178.

[0198] In another aspect, an antibody of the invention comprises (a) a VH domain comprising at least one, at least two, or all three VH HVR sequences selected from (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:176, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:33; and (b) a VL domain comprising at least one, at least two, or all three VL HVR sequences selected from (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37, (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:177, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:178.

[0199] In another aspect, the invention provides an antibody comprising (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:29; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:176; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:177; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:178.

[0200] In any of the above embodiments, an anti-OX40 agonist antibody is humanized. In one embodiment, an anti-OX40 antibody comprises HVRs as in any of the above embodiments or for any of the embodiments in Figure 11, and further comprises an acceptor human framework, e.g. a human immunoglobulin framework or a human consensus framework. In another embodiment, an anti-OX40 antibody comprises HVRs as in any of the above embodiments, and further comprises a VH and/or VL comprising an FR sequence shown in Figure 11.

[0201] In another aspect, an anti-human OX40 agonist antibody comprises a heavy chain variable domain (VH) sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:56, 58, 60, 62, 64, 66, 68, 70, 72, 74, 76, 78, 80, 82, 84, 86, 88, 90, 92, 94, 96, 98, 100, 108, 114 or 116. In certain embodiments, a VH sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO:56, 58, 60, 62, 64, 66, 68, 70, 72, 74, 76, 78, 80, 82, 84, 86, 88, 90, 92, 94, 96,

98, 100, 108, 114 or 116. In certain embodiments, substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VH sequence in SEQ ID NO: SEQ ID NO:56, 58, 60, 62, 64, 66, 68, 70, 72, 74, 76, 78, 80, 82, 84, 86, 88, 90, 92, 94, 96, 98, 100, 108, 114 or 116, including post-translational modifications of that sequence. In a particular embodiment, the VH comprises one, two or three HVRs selected from: (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4.

[0202] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a light chain variable domain (VL) having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:57, 59, 61, 63, 65, 67, 69, 71, 73, 75, 77, 79, 81, 83, 85, 87, 89, 91, 93, 95, 97, 99, 101, 109, 115 or 117. In certain embodiments, a VL sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO: 57, 59, 61, 63, 65, 67, 69, 71, 73, 75, 77, 79, 81, 83, 85, 87, 89, 91, 93, 95, 97, 99, 101, 109, 115 or 117. In certain embodiments, the substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VL sequence in SEQ ID NO: 57, 59, 61, 63, 65, 67, 69, 71, 73, 75, 77, 79, 81, 83, 85, 87, 89, 91, 93, 95, 97, 99, 101, 109, 115 or 117, including post-translational modifications of that sequence. In a particular embodiment, the VL comprises one, two or three HVRs selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7.

[0203] In another aspect, an anti-human OX40 agonist antibody comprises a heavy chain variable domain (VH) sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:56. In certain embodiments, a VH sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in

SEQ ID NO:56. In certain embodiments, substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VH sequence in SEQ ID NO:56, including post-translational modifications of that sequence. In a particular embodiment, the VH comprises one, two or three HVRs selected from: (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4.

[0204] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a light chain variable domain (VL) having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:57. In certain embodiments, a VL sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO: 57. In certain embodiments, the substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VL sequence in SEQ ID NO: 57, including post-translational modifications of that sequence. In a particular embodiment, the VL comprises one, two or three HVRs selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:7.

[0205] In another aspect, an anti-human OX40 agonist antibody comprises a heavy chain variable domain (VH) sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:94. In certain embodiments, a VH sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO:94. In certain embodiments, substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VH sequence in SEQ ID NO:94, including post-translational modifications of that sequence. In a particular embodiment, the VH comprises one, two or

three HVRs selected from: (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4.

[0206] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a light chain variable domain (VL) having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:95. In certain embodiments, a VL sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO:95. In certain embodiments, the substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VL sequence in SEQ ID NO:95, including post-translational modifications of that sequence. In a particular embodiment, the VL comprises one, two or three HVRs selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:26.

[0207] In another aspect, an anti-human OX40 agonist antibody comprises a heavy chain variable domain (VH) sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:96. In certain embodiments, a VH sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO:96. In certain embodiments, substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VH sequence in SEQ ID NO:96, including post-translational modifications of that sequence. In a particular embodiment, the VH comprises one, two or three HVRs selected from: (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4.

[0208] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a light chain variable domain (VL) having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO:97. In certain embodiments, a VL sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO:97. In certain embodiments, the substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VL sequence in SEQ ID NO:97, including post-translational modifications of that sequence. In a particular embodiment, the VL comprises one, two or three HVRs selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:27.

[0209] In another aspect, an anti-human OX40 agonist antibody comprises a heavy chain variable domain (VH) sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO: 118, 120, 122, 124, 126, 128, 130, 132, 134, 136, 138, 140, 142, 144, 146, 148. In certain embodiments, a VH sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO: 118, 120, 122, 124, 126, 128, 130, 132, 134, 136, 138, 140, 142, 144, 146, 148. In certain embodiments, substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VH sequence in SEQ ID NO: SEQ ID NO: 118, 120, 122, 124, 126, 128, 130, 132, 134, 136, 138, 140, 142, 144, 146, 148, including post-translational modifications of that sequence. In a particular embodiment, the VH comprises one, two or three HVRs selected from: (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO: 29, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:30, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:33.

[0210] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a light chain variable domain (VL) having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to the amino acid sequence of SEQ ID NO: 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, 139, 141, 143, 145, 147, 149. In certain embodiments, a VL sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity contains substitutions (e.g., conservative substitutions), insertions, or deletions relative to the reference sequence, but an anti-human OX40 agonist antibody comprising that sequence retains the ability to bind to OX40. In certain embodiments, a total of 1 to 10 amino acids have been substituted, inserted and/or deleted in SEQ ID NO: 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, 139, 141, 143, 145, 147, 149. In certain embodiments, the substitutions, insertions, or deletions occur in regions outside the HVRs (i.e., in the FRs). Optionally, the anti-human OX40 agonist antibody comprises the VL sequence in SEQ ID NO: 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, 139, 141, 143, 145, 147, 149, including post-translational modifications of that sequence. In a particular embodiment, the VL comprises one, two or three HVRs selected from (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:37; (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:39; and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:42.

[0211] In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:56 and SEQ ID NO:57, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:58 and SEQ ID NO:59, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:60 and SEQ ID NO:61, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:62 and SEQ ID NO:63, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:64 and SEQ ID NO:65, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:66 and SEQ ID NO:67, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:68 and SEQ ID NO:69, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:70 and SEQ ID NO:71, respectively, including post-translational modifications of those

sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:72 and SEQ ID NO:73, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:74 and SEQ ID NO:75, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:76 and SEQ ID NO:77, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:78 and SEQ ID NO:79, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:80 and SEQ ID NO:81, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:82 and SEQ ID NO:83, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:84 and SEQ ID NO:85, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:86 and SEQ ID NO:87, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:88 and SEQ ID NO:89, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:90 and SEQ ID NO:91, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:92 and SEQ ID NO:93, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:94 and SEQ ID NO:95, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:96 and SEQ ID NO:97, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:98 and SEQ ID NO:99, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:100 and SEQ ID NO:101, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:108 and SEQ ID NO:109, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:114 and SEQ ID NO:115, respectively, including post-

translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:116 and SEQ ID NO:117, respectively, including post-translational modifications of those sequences.

[0212] In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:118 and SEQ ID NO:119, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:120 and SEQ ID NO:121, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:122 and SEQ ID NO:123, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:124 and SEQ ID NO:125, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:126 and SEQ ID NO:127, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:128 and SEQ ID NO:129, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:130 and SEQ ID NO:131, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:132 and SEQ ID NO:133, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:134 and SEQ ID NO:135, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:136 and SEQ ID NO:137, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:138 and SEQ ID NO:139, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:140 and SEQ ID NO:141, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:142 and SEQ ID NO:143, respectively, including post-translational modifications of those sequences. In one embodiment, the antibody comprises the VH and VL sequences in SEQ ID NO:144 and SEQ ID NO:145, respectively, including post-translational modifications of those sequences. In one embodiment, the

antibody comprises the VH and VL sequences in SEQ ID NO:146 and SEQ ID NO:147, respectively, including post-translational modifications of those sequences.

[0213] In another aspect, an anti-human OX40 agonist antibody is provided, wherein the antibody comprises a VH as in any of the embodiments provided above, and a VL as in any of the embodiments provided above.

[0214] In some embodiments, the OX40 agonist antibody is MEDI6469. In some embodiments, the OX40 agonist antibody is MEDI0562.

[0215] In a further aspect, the invention provides an antibody that binds to the same epitope as an anti-human OX40 antibody provided herein. In some embodiments, the antibody is an anti-human OX40 agonist antibody.

[0216] In a further aspect of the invention, an anti-OX40 antibody according to any of the above embodiments is a monoclonal antibody, including a chimeric, humanized or human antibody. In one embodiment, an anti-OX40 antibody is an antibody fragment, e.g., a Fv, Fab, Fab', scFv, diabody, or F(ab')2 fragment. In another embodiment, the antibody is a full length antibody, e.g., an intact IgG1 antibody or other antibody class or isotype as defined herein. In some embodiments, the antibody is a full length intact IgG4 antibody.

[0217] Exemplary amino acid sequences corresponding to OX40 polypeptides and OX40 antibodies are provided below.

Table 2. Amino acid sequences

Name	SEQUENCE	SEQ ID NO:
Human OX40 (lacking the signal peptide)	LHCVGDTYPSNDRCCHECRPGNGMVSRCSRSQNTVCRPCGPG FYNDVVSSKPCKPCTWCNLRSGSERKQLCTATQDTVCRCRAG TQPLDSYKPGVDCAPCPPGHFSPGDNQACKPWTNCTLAGKHT LQPASNSSDAICEDRDPPATQPQETQGPPARPITVQPTEAWPRT SQGPSTRPVEVPGGRAVAAILGLGLVLGLLGPLAILLALYLLRR DQRLPPDAHKPPGGGSFRTPIQEEQADAHSTLAKI	1
HVR-H1- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5 1A7.gr.6 1A7.gr.7 1A7.gr.NADS 1A7.gr.NADA 1A7.gr.NGDA 1A7.gr.SGDS	DSYMS	2

1A7.gr.NGSS 1A7.Ala.1 1A7.Ala.2 1A7.Ala.3 1A7.Ala.4 1A7.Ala.5 1A7.Ala.6 1A7.Ala.7 1A7.Ala.8 1A7.Ala.9 1A7.Ala.10 1A7.Ala.11 1A7.Ala.12 1A7.Ala.13 1A7.Ala.14 1A7.Ala.15 1A7.Ala.16		
HVR-H2- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5 1A7.gr.6 1A7.gr.7 1A7.gr.DA 1A7.gr.ES 1A7.Ala.1 1A7.Ala.2 1A7.Ala.3 1A7.Ala.4 1A7.Ala.5 1A7.Ala.6 1A7.Ala.7 1A7.Ala.8 1A7.Ala.9 1A7.Ala.10 1A7.Ala.11 1A7.Ala.12 1A7.Ala.13 1A7.Ala.14 1A7.Ala.15 1A7.Ala.16	DMYPDNGDSSYNQKFRE	3
HVR-H3- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5	APRWYFSV	4

1A7.gr.6 1A7.gr.7 1A7.gr.DA 1A7.gr.ES 1A7.gr.NADS 1A7.gr.NADA 1A7.gr.NGDA 1A7.gr.SGDS 1A7.gr.NGSS 1A7.gr.DANA DA 1A7.Ala.1 1A7.Ala.2 1A7.Ala.3 1A7.Ala.4 1A7.Ala.5 1A7.Ala.6 1A7.Ala.7 1A7-Ala.15 1A7.Ala.16		
HVR-L1- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5 1A7.gr.6 1A7.gr.7 1A7.gr.DA 1A7.gr.ES 1A7.gr.NADS 1A7.gr.NADA 1A7.gr.NGDA 1A7.gr.SGDS 1A7.gr.NGSS 1A7.gr.DANA DA 1A7.Ala.1 1A7.Ala.2 1A7.Ala.3 1A7.Ala.4 1A7.Ala.5 1A7.Ala.6 1A7.Ala.7 1A7.Ala.8 1A7.Ala.9 1A7.Ala.10 1A7.Ala.11 1A7.Ala.12 1A7.Ala.13 1A7.Ala.14	RASQDISNYLN	5

1A7.Ala.15 1A7.Ala.16		
HVR-L2- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5 1A7.gr.6 1A7.gr.7 1A7.gr.DA 1A7.gr.ES 1A7.gr.NADS 1A7.gr.NADA 1A7.gr.NGDA 1A7.gr.SGDS 1A7.gr.NGSS 1A7.gr.DANA DA 1A7.Ala.1 1A7.Ala.2 1A7.Ala.3 1A7.Ala.4 1A7.Ala.5 1A7.Ala.6 1A7.Ala.7 1A7.Ala.8 1A7.Ala.9 1A7.Ala.10 1A7.Ala.11 1A7.Ala.12 1A7.Ala.13 1A7.Ala.14 1A7.Ala.15 1A7.Ala.16	YTSRLRS	6
HVR-L3- 1A7.gr.1 1A7.gr.2 1A7.gr.3 1A7.gr.4 1A7.gr.5 1A7.gr.6 1A7.gr.7 1A7.gr.DA 1A7.gr.ES 1A7.gr.NADS 1A7.gr.NADA 1A7.gr.NGDA 1A7.gr.SGDS	QQGHTLPPT	7

1A7.gr.NGSS 1A7.gr.DANA DA 1A7.Ala.8 1A7.Ala.9 1A7.Ala.10 1A7.Ala.11 1A7.Ala.12 1A7.Ala.13 1A7.Ala.14 1A7.Ala.15 1A7.Ala.16		
HVR-H1- 1A7.gr.DA	DAYMS	8
HVR-H1- 1A7.gr.ES 1A7.gr.DANA DA	ESYMS	9
HVR-H2- 1A7.gr.NADS	DMYPDNADSSYNQKFRE	10
HVR-H2- 1A7.gr.NADA 1A7.gr.DANA DA	DMYPDNADASYNQKFRE	11
HVR-H2- 1A7.gr.NGDA	DMYPDNGDASYNQKFRE	12
HVR-H2- 1A7.gr.SGDS	DMYPDSGDSSYNQKFRE	13
HVR-H2- 1A7.gr.NGSS	DMYPDNGSSSYNQKFRE	14
HVR-H3- 1A7.Ala.8	APRWYFSA	15
HVR-H3- 1A7.Ala.9	APRWYASV	16
HVR-H3- 1A7.Ala.10	APRWAFSV	17
HVR-H3- 1A7.Ala.11	APAWYFSV	18
HVR-H3- 1A7.Ala.12	APRWYFAV	19

HVR-H3-1A7.Ala.13	APRAYFSV	20
HVR-H3-1A7.Ala.14	AARWYFSV	21
HVR-L3-1A7.Ala.1	QQGHTLPAT	22
HVR-L3-1A7.Ala.2	QQGHTAPPT	23
HVR-L3-1A7.Ala.3	QQGATLPPT	24
HVR-L3-1A7.Ala.4	QQGHALPPT	25
HVR-L3-1A7.Ala.5	QQAHTLPPT	26
HVR-L3-1A7.Ala.6	QQGHTLAPT	27
HVR-L3-1A7.Ala.7	QAGHTLPPT	28
HVR-H1-3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.SG 3C8.gr.5.EG 3C8.gr.5.QG 3C9.gr.5.DQ 3C8.gr.5.DA 3C8.gr.6 3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11 3C8.A.1 3C8.A.2 3C8.A.3	NYLIE	29

3C8.A.4 3C8.A.5 3C8.A.6 3C8.A.7 3C8.A.8 3C8.A.9 3C8.A.10		
HVR-H2- 3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.SG 3C8.gr.5.EG 3C8.gr.5.QG 3C8.gr.6 3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11 3C8.A.1 3C8.A.2 3C8.A.3 3C8.A.4 3C8.A.5 3C8.A.6 3C8.A.7 3C8.A.8 3C8.A.9 3C8.A.10	VINPGSGDTYYSEKFKG	30
HVR-H2- 3C8.gr.5.DA	VINPGSGDAYYSEKFKG	31
HVR-H2- 3C8.gr.5.DQ	VINPGSGDQYYSEKFKG	32
HVR-H3- 3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.SG 3C8.gr.5.EG 3C8.gr.5.QG 3C8.gr.5.DA 3C8.gr.5.DQ 3C8.gr.6	DRLDY	33

3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11 3C8.A.1 3C8.A.2 3C8.A.3 3C8.A.4 3C8.A.5 3C8.A.6 3C8.A.7		
HVR-H3- 3C8.A.8	ARLDY	34
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HVR-L1- 3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.SG 3C8.gr.5.EG 3C8.gr.5.QG 3C8.gr.5.DA 3C8.gr.5.DQ 3C8.gr.6 3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11 3C8.A.1 3C8.A.2 3C8.A.3 3C8.A.4 3C8.A.5 3C8.A.6 3C8.A.7 3C8.A.8 3C8.A.9 3C8.A.10	HASQDISSYIV	37
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3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.DA 3C8.gr.5.DQ 3C8.gr.6 3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11 3C8.A.1 3C8.A.2 3C8.A.3 3C8.A.4 3C8.A.5 3C8.A.6 3C8.A.7 3C8.A.8 3C8.A.9 3C8.A.10		
HVR-L2- 3C8.gr5.SG	HGTNLES	39
HVR-L2- 3C8.gr.5.EG	HGTNLEE	40
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HVR-L3 3C8.gr.1 3C8.gr.2 3C8.gr.3 3C8.gr.4 3C8.gr.5 3C8.gr.5.SG 3C8.gr.5.EG 3C8.gr.5.QG 3C8.gr.5.DA 3C8.gr.5.DQ 3C8.gr.6 3C8.gr.7 3C8.gr.8 3C8.gr.9 3C8.gr.10 3C8.gr.11	VHYAQFPYT	42

3C8.A.8 3C8.A.9 3C8.A.10		
HVR-L3- 3C8.A.1	AHYAQFPYT	43
HVR-L3- 3C8.A.2	VAYAQFPYT	44
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HVR-L3- 3C8.A.4	VHYAAFPYT	46
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HVR-L3- 3C8.A.6	VHYAQFAYT	48
HVR-L3- 3C8.A.7	VHYAQFPAT	49
HVR-H1- 1D2.gr.1 1D2.gr.2 1D2.gr.3	DYGVL	50
HVR-H2- 1D2.gr.1 1D2.gr.2 1D2.gr.3	MIWSGGTTDYNAAFIS	51
HVR-H3- 1D2.gr.1 1D2.gr.2 1D2.gr.3	EEMDY	52
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HVR-L3- 1D2.gr.1 1D2.gr.2 1D2.gr.3	QQGNTLPWT	55
1A7.gr.1	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP	56

V_H	GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	
1A7.gr.1 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	57
1A7.gr.2 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITVDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	58
1A7.gr.2 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	59
1A7.gr.3 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTLTVDTSTSTAYLEL SSLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	60
1A7.gr.3 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	61
1A7.gr.4 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITVDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	62
1A7.gr.4 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKT VKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	63
1A7.gr.5 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITVDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	64
1A7.gr.5 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKT VKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	65
1A7.gr.6 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITVDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	66
1A7.gr.6 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKT VKLLIYYTSRLRSGVPSRFSGSGSGKDYTLTISSLQPEDFATYFC QQGHTLPPTFGQGTKVEIK	67
1A7.gr.7 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITVDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	68
1A7.gr.7 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKT VKLLIYYTSRLRSGVPSRFSGSGSGKDYTLTISSLQPEDFATYFC QQGHTLPPTFGQGTKVEIK	69
1A7.gr.DA V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDAYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	70
1A7.gr.DA V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	71
1A7.gr.ES V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTESYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	72

1A7.gr.ES V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	73
1A7.gr.NADS V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNADSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	74
1A7.gr.NADS V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	75
1A7.gr.NADA V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNADASYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	76
1A7.gr.NADA V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	77
1A7.gr.NGDA V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDASYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	78
1A7.gr.NGDA V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	79
1A7.gr.SGDS V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDSGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	80
1A7.gr.SGDS V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	81
1A7.gr.NGSS V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGSSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	82
1A7.gr.NGSS V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	83
1A7.gr.DANA DA V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDAYMSWVRQAP GQGLEWIGDMYPDNADASYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	84
1A7.gr.DANA DA V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	85
1A7.Ala.1 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	86
1A7.Ala.1 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPATFGQGTKVEIK	87
1A7.Ala.2 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	88
1A7.Ala.2 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC	89

	QQGHTAPPTFGQGTKVEIK	
1A7.Ala.3 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	90
1A7.Ala.3 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGATLPPTFGQGTKVEIK	91
1A7.Ala.4 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	92
1A7.Ala.4 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHALPPTFGQGTKVEIK	93
1A7.Ala.5 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	94
1A7.Ala.5 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQAHTLPPTFGQGTKVEIK	95
1A7.Ala.6 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	96
1A7.Ala.6 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLAPTFGQGTKVEIK	97
1A7.Ala.7 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	98
1A7.Ala.7 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QAGHTLPPTFGQGTKVEIK	99
1A7.Ala.8 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFSAWGQGTLVTVSS	100
1A7.Ala.8 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	101
1A7.Ala.9 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYASVWGQGTLVTVSS	102
1A7.Ala.9 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	103
1A7.Ala.10 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWAFSVWGQGTLVTVSS	104
1A7.Ala.10 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	105
1A7.Ala.11	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP	106

V_H	GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPAWYFSVWGQGTLVTVSS	
1A7.Ala.11 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	107
1A7.Ala.12 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRWYFAVWGQGTLVTVSS	108
1A7.Ala.12 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	109
1A7.Ala.13 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAPRAYFSVWGQGTLVTVSS	110
1A7.Ala.13 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	111
1A7.Ala.14 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVLAARWYFSVWGQGTLVTVSS	112
1A7.Ala.14 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	113
1A7.Ala.15 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCALAPRWYFSVWGQGTLVTVSS	114
1A7.Ala.15 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	115
1A7.Ala.16 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAP GQGLEWIGDMYPDNGDSSYNQKFRERVTITRDTSTSTAYLELS SLRSEDTAVYYCVAAPRWYFSVWGQGTLVTVSS	116
1A7.Ala.16 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKA PKLLIYYTSRLRSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGHTLPPTFGQGTKVEIK	117
3C8.gr.1 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTITRDTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	118
3C8.gr.1 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKAP KLLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	119
3C8.gr.2 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTITADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	120
3C8.gr.2 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKAP KLLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	121
3C8.gr.3 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL	122

	RSEDTAVYYCARDRLDYWGQGTLVTVSS	
3C8.gr.3 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKAP KLLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	123
3C8.gr.4 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTITADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	124
3C8.gr.4 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	125
3C8.gr.5 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	126
3C8.gr.5 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	127
3C8.gr.5.SG V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	128
3C8.gr.5.SG V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLESGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	129
3C8.gr.5.EG V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	130
3C8.gr.5.EG V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEEGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	131
3C8.gr.5.QG V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	132
3C8.gr.5.QG V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEQGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	133
3C8.gr.6 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTITADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	134
3C8.gr.6 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGADYTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	135
3C8.gr.7 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	136
3C8.gr.7 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGADYTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	137
3C8.gr.8 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTRDTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	138
3C8.gr.8	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF	139

V_L	KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	
3C8.gr.9 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTRDTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	140
3C8.gr.9 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSP KLLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	141
3C8.gr.10 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTRDTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	142
3C8.gr.10 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKAF KLLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	143
3C8.gr.11 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTRDTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	144
3C8.gr.11 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKAP KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	145
3C8.A.1 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	146
3C8.A.1 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC AHYAQFPYTFGQGTKVEIK	147
3C8.A.2 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	148
3C8.A.2 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VAYAQFPYTFGQGTKVEIK	149
3C8.A.3 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	150
3C8.A.3 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHAAQFPYTFGQGTKVEIK	151
3C8.A.4 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	152
3C8.A.4 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAAFPYTFGQGTKVEIK	153
3C8.A.5 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	154
3C8.A.5	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF	155

V_L	KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQAPYTFGQGTKVEIK	
3C8.A.6 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	156
3C8.A.6 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFAYTFGQGTKVEIK	157
3C8.A.7 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRLDYWGQGTLVTVSS	158
3C8.A.7 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPATFGQGTKVEIK	159
3C8.A.8 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARARLDYWGQGTLVTVSS	160
3C8.A.8 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	161
3C8.A.9 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDALDYWGQGTLVTVSS	162
3C8.A.9 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	163
3C8.A.10 V_H	EVQLVQSGAEVKKPGASVKVSCKASGYAFTNYLIEWVRQAPG QGLEWIGVINPGSGDTYYSEKFKGRVTLTADTSTSTAYLELSSL RSEDTAVYYCARDRADYWGQGTLVTVSS	164
3C8.A.10 V_L	DIQMTQSPSSLSASVGDRVTITCHASQDISSYIVWYQQKPGKSF KGLIYHGTNLEDGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC VHYAQFPYTFGQGTKVEIK	165
1D2.gr.1 V_H	EVQLVESGPGLVKPSETLSLTCTVSGFSLTDYGVLWIRQPPGK GLEWIGMIWSGGTTDYNAAFISRVTISVDTSKNQFSLKLSSVTA ADTAVYYCVREEMDYWGQGTLVTVSS	166
1D2.gr.1 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNFLNWYQQKPGKA PKLLIYYTSRLHSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGNTLPWTFGQGTKVEIK	167
1D2.gr.2 V_H	EVQLVESGPGLVKPSETLSLTCTVSGFSLTDYGVLWIRQPPGK GLEWIGMIWSGGTTDYNAAFISRVTISKDTSKNQVSLKLSSVT AADTAVYYCVREEMDYWGQGTLVTVSS	168
1D2.gr.2 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNFLNWYQQKPGKA PKLLIYYTSRLHSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQGNTLPWTFGQGTKVEIK	169
1D2.gr.3 V_H	EVQLVESGPGLVKPSETLSLTCTVSGFSLTDYGVLWVRQPPGK GLEWLGMIWSGGTTDYNAAFISRLTISKDTSKNQVSLKLSSVT AADTAVYYCVREEMDYWGQGTLVTVSS	170
1D2.gr.3 V_L	DIQMTQSPSSLSASVGDRVTITCRASQDISNFLNWYQQKPGKA PKLLIYYTSRLHSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYC	171

	QQGNTLPWTFGQGTKVEIK	
CON1 (1A7)HVR-H1	X_1X_2YMS, wherein X_1 is D or E, and X_2 is S or A	172
CON1 (1A7) HVR-H2	DMYPD$X_1X_2X_3X_4$SYNQKFRE, wherein X_1 is N or S, X_1 is A or G, X_3 is D or S, and X_4 is A or S	173
CON1 (1A7) HVR-H3	APRW$X_1X_2X_3X_4$, wherein X_1 is Y or A, X_2 is A or F, X_3 is S or A, and X_4 is A or V.	174
CON1 (1A7) HVR-L3	Q$X_1X_2X_3X_4X_5X_6X_7$T, wherein X_1 is A or Q, X_2 is A or G, X_3 is A or H, X_4 is A or T, X_5 is A or L, X_6 is A or P, and X_7 is A or P.	175
CON2 (3C8) HVR-H2	VINPGSGDX_1YYSEKFKG, wherein X_1 is T, A or Q.	176
CON2 (3C8) HVR-L2	HGTNLEX_1, wherein X_1 is S, E, or Q.	177
CON2 (3C8) HVR-L3	X_1X_2YAQFPYX_3, wherein X_1 is V or A, X_2 is H or A, and X_3 is Y or A.	178
1A7 V_L	DIQMTQTTSSLSASLGDRVTISCRASQDISNYLNWYQQKPDGTVKLLIYYTSRLRSGVPSRFSGSGSGKDYFLTISNLEQEDVAAYFCQQGHTLPPTFGGGTKLEIK	179
1A7 V_H	EVQLQQSGPELVKPGASVKISCKASGYTFTDSYMSWVKQSHGKTLEWIGDMYPDNGDSSYNQKFREKVTLTVDKSSTTAYMEFRSLTSEDSAVYYCVLAPRWYFSVWGTGTTVTVSS	180
3C8 V_L	DILMTQSPSSMSVSLGDTVSITCHASQDISSYIVWLQQKPGKSFRGLIYHGTNLEDGIPSRFSGSGSGADYSLTISSLESEDFADYYCVHYAQFPYTFGGGTKLEIK	181
3C8 V_H	QVQLQQSGAELVRPGTSVKVSCKASGYAFTNYLIEWVKQRPGQGLEWIGVINPGSGDTYYSEKFKGKVTLTADKSSSTAYMQLSSLTSEDSAVYFCARDRLDYWGQGTTLTVSS	182
1A7.gr.5' V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAPGQGLEWIGDMYPDNGDSSYNQKFRERVTLTVDTSTSTAYLELSSLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	225
1A7.gr.7' V_H	EVQLVQSGAEVKKPGASVKVSCKASGYTFTDSYMSWVRQAPGQGLEWIGDMYPDNGDSSYNQKFRERVTLTVDTSTSTAYLELSSLRSEDTAVYYCVLAPRWYFSVWGQGTLVTVSS	226

[0218] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in US7550140. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain comprising the sequence of EVQLVESGGGLVQPGGSLRLSCAASGFTFSNYTMNWVRQAPGKGLEWVSAISGSGGSTYYADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCAKDRYSQVHYALDYWGQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKT

TPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK (SEQ ID NO:183)
and/or a light chain comprising the sequence of
DIVMTQSPDSLPVTPGEPASISCRSSQSLLHSNGYNYLDWYLQKAGQSPQLLIYLGSN RASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCQQYYNHPTTFGQGTKLEIKRTV AAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQD SKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO:184). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody 008 as described in US7550140. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody 008 as described in US7550140.

[0219] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in US7550140. In some embodiments, the agonist anti-human OX40 antibody comprises the sequence of
MAEVQLVESGGGLVQPGGSLRLSCAASGFTFSNYTMNWVRQAPGKGLEWVSAISGS GGSTYYADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCAKDRYSQVHYALD YWGQGTLVTVLEGTGGSGGTGSGTGTSELDIQMTQSPDSLPVTPGEPASISCRSSQSL LHSNGYNYLDWYLQKAGQSPQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAE DVGVYYCQQYYNHPTTFGQGTKLEIKRAA (SEQ ID NO:185). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody SC02008 as described in US7550140. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody SC02008 as described in US7550140.

[0220] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in US7550140. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain comprising the sequence of
EVQLVESGGGLVHPGGSLRLSCAGSGFTFSSYAMHWVRQAPGKGLEWVSAIGTGGG TYYADSVMGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCARYDNVMGLYWFDYW GQGTLVTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALT SGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDK THTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVD GVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTI SKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKT

TPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK (SEQ ID NO:186) and/or a light chain comprising the sequence of EIVLTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDASNRATGIPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQRSNWPPAFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO:187). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody 023 as described in US7550140. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody 023 as described in US7550140.

[0221] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in US7960515. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of EVQLVESGGGLVQPGGSLRLSCAASGFTFSSYSMNWVRQAPGKGLEWVSYISSSSSTIDYADSVKGRFTISRDNAKNSLYLQMNSLRDEDTAVYYCARESGWYLFDYWGQGTLVTVSS (SEQ ID NO:188) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQGISSWLAWYQQKPEKAPKSLIYAASSLQSGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYNSYPPTFGGGTKVEIK (SEQ ID NO:189). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody 11D4 as described in US7960515. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody 11D4 as described in US7960515.

[0222] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in US7960515. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain comprising the sequence of EVQLVESGGGLVQPGRSLRLSCAASGFTFDDYAMHWVRQAPGKGLEWVSGISWNSGSIGYADSVKGRFTISRDNAKNSLYLQMNSLRAEDTALYYCAKDQSTADYYFYYGMDVWGQGTTVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK (SEQ ID NO:190) and/or a light chain comprising the sequence of

EIVVTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDASNRATGI PARFSGSGSGTDFTLTISSLEPEDFAVYYCQQRSNWPTFGQGTKVEIKRTVAAPSVFIF PPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYS LSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO:191). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody 18D8 as described in US7960515. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody 18D8 as described in US7960515.

[0223] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2012/027328. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGSELKKPGASVKVSCKASGYTFTDYSMHWVRQAPGQGLKWMGWINTE TGEPTYADDFKGRFVFSLDTSVSTAYLQISSLKAEDTAVYYCANPYYDYVSYYAMD YWGQGTTVTVSS (SEQ ID NO:192) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVSTAVAWYQQKPGKAPKLLIYSASYLYTG VPSRFSGSGSGTDFTFTISSLQPEDIATYYCQQHYSTPRTFGQGTKLEIK (SEQ ID NO:193). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody hu106-222 as described in WO2012/027328. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody hu106-222 as described in WO2012/027328.

[0224] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2012/027328. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of EVQLVESGGGLVQPGGSLRLSCAASEYEFPSHDMSWVRQAPGKGLELVAAINSDGG STYYPDTMERRFTISRDNAKNSLYLQMNSLRAEDTAVYYCARHYDDYYAWFAYWG QGTMVTVSS (SEQ ID NO:194) and/or a light chain variable region comprising the sequence of EIVLTQSPATLSLSPGERATLSCRASKSVSTSGYSYMHWYQQKPGQAPRLLIYLASNL ESGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQHSRELPLTFGGGTKVEIK (SEQ ID NO:195). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody Hu119-122 as described in WO2012/027328. In some embodiments, the antibody comprises a heavy chain variable

region sequence and/or a light chain variable region sequence of antibody Hu119-122 as described in WO2012/027328.

[0225] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2013/028231. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain comprising the sequence of MYLGLNYVFIVFLLNGVQSEVKLEESGGGLVQPGGSMKLSCAASGFTFSDAWMDW VRQSPEKGLEWVaEIRSKANNHATYYAESVNGRFTISRDDSKSSVYLQMNSLRAEDT GIYYCTWGEVFYFDYWGQGTTLTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKD YFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTYITCNVNHK PSNTKVDKKVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVV VDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGK EYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPS DIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEA LHNHYTQKSLSLSPGK (SEQ ID NO:196) and/or a light chain comprising the sequence of MRPSIQFLGLLLFWLHGAQCDIQMTQSPSSLSASLGGKVTITCKSSQDINKYIAWYQH KPGKGPRLLIHYTSTLQPGIPSRFSGSGSGRDYSFSISNLEPEDIATYYCLQYDNLLTFG AGTKLELKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGE C (SEQ ID NO:197). In some embodiments, the anti-human OX40 agonist antibody comprises a heavy chain variable region comprising the sequence of MYLGLNYVFIVFLLNGVQSEVKLEESGGGLVQPGGSMKLSCAASGFTFSDAWMDWVRQSPE KGLEWVAEIRSKANNHATYYAESVNGRFTISRDDSKSSVYLQMNSLRAEDTGIYYCTWGEV FYFDYWGQGTTLTVSS (SEQ ID NO:198) and/or a light chain variable region comprising the sequence of MRPSIQFLGLLLFWLHGAQCDIQMTQSPSSLSASLGGKVTITCKSSQDINKYIAWYQHKPGKG PRLLIHYTSTLQPGIPSRFSGSGSGRDYSFSISNLEPEDIATYYCLQYDNLLTFGAGTKLELK (SEQ ID NO:199). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody Mab CH 119-43-1 as described in WO2013/028231. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody Mab CH 119-43-1 as described in WO2013/028231.

[0226] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2013038191. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of EVQLQQSGPELVKPGASVKMSCKASGYTFTSYVMHWVKQKPGQGLEWIGYINPYN

DGTKYNEKFKGKATLTSDKSSSTAYMELSSLTSEDSAVYYCANYYGSSLSMDYWG QGTSVTVSS (SEQ ID NO:200) and/or a light chain variable region comprising the sequence of DIQMTQTTSSLSASLGDRVTISCRASQDISNYLNWYQQKPDGTVKLLIYYTSRLHSGV PSRFSGSGSGTDYSLTISNLEQEDIATYFCQQGNTLPWTFGGGTKLEIKR (SEQ ID NO:201). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2013038191. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2013038191.

[0227] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2013038191. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of EVQLQQSGPELVKPGASVKISCKTSGYTFKDYTMHWVKQSHGKSLEWIGGIYPNNG GSTYNQNFKDKATLTVDKSSSTAYMEFRSLTSEDSAVYYCARMGYHGPHLDFDVW GAGTTVTVSP (SEQ ID NO:202) and/or a light chain variable region comprising the sequence of DIVMTQSHKFMSTSLGDRVSITCKASQDVGAAVaWYQQKPGQSPKLLIYWASTRHT GVPDRFTGGGSGTDFTLTISNVQSEDLTDYFCQQYINYPLTFGGGTKLEIKR (SEQ ID NO:203). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2013038191. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2013038191.

[0228] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWMGYINPY NDGTKYNEKFKGRVTITSDTSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWG QGTLVTVSS (SEQ ID NO:204) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAPKLLIYYTSRLHSGV PSRFSGSGSGTDYTLTISSLQPEDFATYYCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:205). In some embodiments, the antibody comprises at least one, two, three, four, five or

six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0229] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWMGYINPY NDGTKYNEKFKGRVTITSDTSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWG QGTLVTVSS (SEQ ID NO:204) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAVKLLIYYTSRLHSG VPSRFSGSGSGTDYTLTISSLQPEDFATYFCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:206). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0230] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWIGYINPYN DGTKYNEKFKGRATITSDTSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWGQ GTLVTVSS (SEQ ID NO:207) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAPKLLIYYTSRLHSGV PSRFSGSGSGTDYTLTISSLQPEDFATYYCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:205). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0231] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human

OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWIGYINPYNDGTKYNEKFKGRATITSDTSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWGQGTLVTVSS (SEQ ID NO:207) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAVKLLIYYTSRLHSGVPSRFSGSGSGTDYTLTISSLQPEDFATYFCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:206). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0232] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWIGYINPYNDGTKYNEKFKGRATLTSDKSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWGQGTLVTVSS (SEQ ID NO:208) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAPKLLIYYTSRLHSGVPSRFSGSGSGTDYTLTISSLQPEDFATYYCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:205). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0233] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYVMHWVRQAPGQRLEWIGYINPYNDGTKYNEKFKGRATLTSDKSASTAYMELSSLRSEDTAVYYCANYYGSSLSMDYWGQGTLVTVSS (SEQ ID NO:208) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCRASQDISNYLNWYQQKPGKAVKLLIYYTSRLHSG

VPSRFSGSGSGTDYTLTISSLQPEDFATYFCQQGNTLPWTFGQGTKVEIKR (SEQ ID NO:206). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 20E5 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 20E5 as described in WO2014148895A1.

[0234] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWMGGIYPN NGGSTYNQNFKDRVTITADKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDV WGQGTTVTVSS (SEQ ID NO:209) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTG VPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:210). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0235] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWMGGIYPN NGGSTYNQNFKDRVTITADKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDV WGQGTTVTVSS (SEQ ID NO:209) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTG VPDRFSGGGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:211). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0236] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWIGGIYPNNGGSTYNQNFKDRVTLTADKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDVWGQGTTVTVSS (SEQ ID NO:212) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:210). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0237] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWIGGIYPNNGGSTYNQNFKDRVTLTADKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDVWGQGTTVTVSS (SEQ ID NO:212) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTGVPDRFSGGGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:211). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0238] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWIGGIYPNNGGSTYNQNFKDRATLTVDKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDVWGQGTTVTVSS (SEQ ID NO:213) and/or a light chain variable region comprising the

sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTG VPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:210). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0239] In some embodiments, the agonist anti-human OX40 antibody is an anti-OX40 antibody described in WO2014148895A1. In some embodiments, the agonist anti-human OX40 antibody comprises a heavy chain variable region comprising the sequence of QVQLVQSGAEVKKPGSSVKVSCKASGYTFKDYTMHWVRQAPGQGLEWIGGIYPNN GGSTYNQNFKDRATLTVDKSTSTAYMELSSLRSEDTAVYYCARMGYHGPHLDFDV WGQGTTVTVSS (SEQ ID NO:213) and/or a light chain variable region comprising the sequence of DIQMTQSPSSLSASVGDRVTITCKASQDVGAAVaWYQQKPGKAPKLLIYWASTRHTG VPDRFSGGGSGTDFTLTISSLQPEDFATYYCQQYINYPLTFGGGTKVEIKR (SEQ ID NO:211). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody clone 12H3 as described in WO2014148895A1. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody clone 12H3 as described in WO2014148895A1.

[0240] In some embodiments, the agonist anti-human OX40 antibody is L106 BD (Pharmingen Product # 340420). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody L106 (BD Pharmingen Pduct # 340420). In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody L106 (BD Pharmingen Product # 340420).

[0241] In some embodiments, the agonist anti-human OX40 antibody is ACT35 (Santa Cruz Biotechnology, Catalog # 20073). In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody ACT35 (Santa Cruz Biotechnology, Catalog # 20073). In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody ACT35 (Santa Cruz Biotechnology, Catalog # 20073).

[0242] In some embodiments, the agonist anti-human OX40 antibody is MEDI6469. In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody MEDI6469. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody MEDI6469.

[0243] In some embodiments, the agonist anti-human OX40 antibody is MEDI0562. In some embodiments, the antibody comprises at least one, two, three, four, five or six hypervariable region (HVR) sequences of antibody MEDI0562. In some embodiments, the antibody comprises a heavy chain variable region sequence and/or a light chain variable region sequence of antibody MEDI0562.

Other OX40 Agonists

[0244] OX40 agonists useful for the methods described herein are in no way intended to be limited to antibodies. Non-antibody OX40 agonists are contemplated and well known in the art.

[0245] As described above, OX40L (also known as CD134L) serves as a ligand for OX40. As such, agonists that present part or all of OX40L may serve as OX40 agonists. In some embodiments, an OX40 agonist may include one or more extracellular domains of OX40L. Examples of extracellular domains of OX40L may include OX40-binding domains. In some embodiments, an OX40 agonist may be a soluble form of OX40L that includes one or more extracellular domains of OX40L but lacks other, insoluble domains of the protein, e.g., transmembrane domains. In some embodiments, an OX40 agonist is a soluble protein that includes one or more extracellular domains of OX40L able to bind OX40L. In some embodiments, an OX40 agonist may be linked to another protein domain, e.g., to increase its effectiveness, half-life, or other desired characteristics. In some embodiments, an OX40 agonist may include one or more extracellular domains of OX40L linked to an immunoglobulin Fc domain.

[0246] In some embodiments, an OX40 agonist may be any one of the OX40 agonists described in U.S. Patent No. 7,696,175.

[0247] In some embodiments, an OX40 agonist may be an oligomeric or multimeric molecule. For example, an OX40 agonist may contain one or more domains (e.g., a leucine zipper domain) that allows proteins to oligomerize. In some embodiments, an OX40 agonist may include one or more extracellular domains of OX40L linked to one or more leucine zipper domains.

[0248] In some embodiments, an OX40 agonist may be any one of the OX40 agonists described in European Patent No. EP0672141 B1.

[0249] In some embodiments, an OX40 agonist may be a trimeric OX40L fusion protein. For example, an OX40 agonist may include one or more extracellular domains of OX40L linked to an immunoglobulin Fc domain and a trimerization domain (including without limitation an isoleucine zipper domain).

[0250] In some embodiments, an OX40 agonist may be any one of the OX40 agonists described in International Publication No. WO2006/121810. In some embodiments, the OX40 agonist is MEDI6383.

[0251] In a further aspect, an anti-OX40 agonist and/or antibody according to any of the above embodiments may incorporate any of the features, singly or in combination, as described below.

1. Antibody Affinity

[0252] In certain embodiments, an antibody provided herein has a dissociation constant (Kd) of ≤ 1μM, ≤ 100 nM, ≤ 10 nM, ≤ 1 nM, ≤ 0.1 nM, ≤ 0.01 nM, or ≤ 0.001 nM (e.g. 10-8 M or less, e.g. from 10-8 M to 10-13 M, e.g., from 10-9 M to 10-13 M).

[0253] In one embodiment, Kd is measured by a radiolabeled antigen binding assay (RIA). In one embodiment, an RIA is performed with the Fab version of an antibody of interest and its antigen. For example, solution binding affinity of Fabs for antigen is measured by equilibrating Fab with a minimal concentration of (125I)-labeled antigen in the presence of a titration series of unlabeled antigen, then capturing bound antigen with an anti-Fab antibody-coated plate (see, e.g., Chen et al., J. Mol. Biol. 293:865-881(1999)). To establish conditions for the assay, MICROTITER® multi-well plates (Thermo Scientific) are coated overnight with 5 μg/ml of a capturing anti-Fab antibody (Cappel Labs) in 50 mM sodium carbonate (pH 9.6), and subsequently blocked with 2% (w/v) bovine serum albumin in PBS for two to five hours at room temperature (approximately 23°C). In a non-adsorbent plate (Nunc #269620), 100 pM or 26 pM [125I]-antigen are mixed with serial dilutions of a Fab of interest (e.g., consistent with assessment of the anti-VEGF antibody, Fab-12, in Presta et al., Cancer Res. 57:4593-4599 (1997)). The Fab of interest is then incubated overnight; however, the incubation may continue for a longer period (e.g., about 65 hours) to ensure that equilibrium is reached. Thereafter, the mixtures are transferred to the capture plate for incubation at room temperature (e.g., for one hour). The solution is then removed and the plate washed eight

times with 0.1% polysorbate 20 (TWEEN-20®) in PBS. When the plates have dried, 150 μl/well of scintillant (MICROSCINT-20 TM; Packard) is added, and the plates are counted on a TOPCOUNT TM gamma counter (Packard) for ten minutes. Concentrations of each Fab that give less than or equal to 20% of maximal binding are chosen for use in competitive binding assays.

[0254] According to another embodiment, Kd is measured using a BIACORE® surface plasmon resonance assay. For example, an assay using a BIACORE®-2000 or a BIACORE ®-3000 (BIAcore, Inc., Piscataway, NJ) is performed at 25°C with immobilized antigen CM5 chips at ~10 response units (RU). In one embodiment, carboxymethylated dextran biosensor chips (CM5, BIACORE, Inc.) are activated with N-ethyl-N'- (3-dimethylaminopropyl)-carbodiimide hydrochloride (EDC) and N-hydroxysuccinimide (NHS) according to the supplier's instructions. Antigen is diluted with 10 mM sodium acetate, pH 4.8, to 5 μg/ml (~0.2 μM) before injection at a flow rate of 5 μl/minute to achieve approximately 10 response units (RU) of coupled protein. Following the injection of antigen, 1 M ethanolamine is injected to block unreacted groups. For kinetics measurements, two-fold serial dilutions of Fab (0.78 nM to 500 nM) are injected in PBS with 0.05% polysorbate 20 (TWEEN-20TM) surfactant (PBST) at 25°C at a flow rate of approximately 25 μl/min. Association rates (kon) and dissociation rates (koff) are calculated using a simple one-to-one Langmuir binding model (BIACORE ® Evaluation Software version 3.2) by simultaneously fitting the association and dissociation sensorgrams. The equilibrium dissociation constant (Kd) is calculated as the ratio koff/kon. See, e.g., Chen et al., J. Mol. Biol. 293:865-881 (1999). If the on-rate exceeds 106 M-1 s-1 by the surface plasmon resonance assay above, then the on-rate can be determined by using a fluorescent quenching technique that measures the increase or decrease in fluorescence emission intensity (excitation = 295 nm; emission = 340 nm, 16 nm band-pass) at 25oC of a 20 nM anti-antigen antibody (Fab form) in PBS, pH 7.2, in the presence of increasing concentrations of antigen as measured in a spectrometer, such as a stop-flow equipped spectrophometer (Aviv Instruments) or a 8000-series SLM-AMINCO TM spectrophotometer (ThermoSpectronic) with a stirred cuvette.

2. Antibody Fragments

[0255] In certain embodiments, an antibody provided herein is an antibody fragment. Antibody fragments include, but are not limited to, Fab, Fab', Fab'-SH, F(ab')2, Fv, and scFv fragments, and other fragments described below. For a review of certain antibody fragments, see Hudson et al. Nat. Med. 9:129-134 (2003). For a review of scFv fragments, see, e.g.,

Pluckthün, in The Pharmacology of Monoclonal Antibodies, vol. 113, Rosenburg and Moore eds., (Springer-Verlag, New York), pp. 269-315 (1994); see also WO 93/16185; and U.S. Patent Nos. 5,571,894 and 5,587,458. For discussion of Fab and F(ab')2 fragments comprising salvage receptor binding epitope residues and having increased in vivo half-life, see U.S. Patent No. 5,869,046.

[0256] Diabodies are antibody fragments with two antigen-binding sites that may be bivalent or bispecific. See, for example, EP 404,097; WO 1993/01161; Hudson et al., Nat. Med. 9:129-134 (2003); and Hollinger et al., Proc. Natl. Acad. Sci. USA 90: 6444-6448 (1993). Triabodies and tetrabodies are also described in Hudson et al., Nat. Med. 9:129-134 (2003).

[0257] Single-domain antibodies are antibody fragments comprising all or a portion of the heavy chain variable domain or all or a portion of the light chain variable domain of an antibody. In certain embodiments, a single-domain antibody is a human single-domain antibody (Domantis, Inc., Waltham, MA; see, e.g., U.S. Patent No. 6,248,516 B1).

[0258] Antibody fragments can be made by various techniques, including but not limited to proteolytic digestion of an intact antibody as well as production by recombinant host cells (e.g. E. coli or phage), as described herein.

3. Chimeric and Humanized Antibodies

[0259] In certain embodiments, an antibody provided herein is a chimeric antibody. Certain chimeric antibodies are described, e.g., in U.S. Patent No. 4,816,567; and Morrison et al., Proc. Natl. Acad. Sci. USA, 81:6851-6855 (1984)). In one example, a chimeric antibody comprises a non-human variable region (e.g., a variable region derived from a mouse, rat, hamster, rabbit, or non-human primate, such as a monkey) and a human constant region. In a further example, a chimeric antibody is a "class switched" antibody in which the class or subclass has been changed from that of the parent antibody. Chimeric antibodies include antigen-binding fragments thereof.

[0260] In certain embodiments, a chimeric antibody is a humanized antibody. Typically, a non-human antibody is humanized to reduce immunogenicity to humans, while retaining the specificity and affinity of the parental non-human antibody. Generally, a humanized antibody comprises one or more variable domains in which HVRs, e.g., CDRs, (or portions thereof) are derived from a non-human antibody, and FRs (or portions thereof) are derived from human antibody sequences. A humanized antibody optionally will also comprise at least a portion of a human constant region. In some embodiments, some FR residues in a

humanized antibody are substituted with corresponding residues from a non-human antibody (e.g., the antibody from which the HVR residues are derived), e.g., to restore or improve antibody specificity or affinity.

[0261] Humanized antibodies and methods of making them are reviewed, e.g., in Almagro and Fransson, Front. Biosci. 13:1619-1633 (2008), and are further described, e.g., in Riechmann et al., Nature 332:323-329 (1988); Queen et al., Proc. Nat'l Acad. Sci. USA 86:10029-10033 (1989); US Patent Nos. 5, 821,337, 7,527,791, 6,982,321, and 7,087,409; Kashmiri et al., Methods 36:25-34 (2005) (describing specificity determining region (SDR) grafting); Padlan, Mol. Immunol. 28:489-498 (1991) (describing "resurfacing"); Dall'Acqua et al., Methods 36:43-60 (2005) (describing "FR shuffling"); and Osbourn et al., Methods 36:61-68 (2005) and Klimka et al., Br. J. Cancer, 83:252-260 (2000) (describing the "guided selection" approach to FR shuffling).

[0262] Human framework regions that may be used for humanization include but are not limited to: framework regions selected using the "best-fit" method (see, e.g., Sims et al. J. Immunol. 151:2296 (1993)); framework regions derived from the consensus sequence of human antibodies of a particular subgroup of light or heavy chain variable regions (see, e.g., Carter et al. Proc. Natl. Acad. Sci. USA, 89:4285 (1992); and Presta et al. J. Immunol., 151:2623 (1993)); human mature (somatically mutated) framework regions or human germline framework regions (see, e.g., Almagro and Fransson, Front. Biosci. 13:1619-1633 (2008)); and framework regions derived from screening FR libraries (see, e.g., Baca et al., J. Biol. Chem. 272:10678-10684 (1997) and Rosok et al., J. Biol. Chem. 271:22611-22618 (1996)).

4. Human Antibodies

[0263] In certain embodiments, an antibody provided herein is a human antibody. Human antibodies can be produced using various techniques known in the art. Human antibodies are described generally in van Dijk and van de Winkel, Curr. Opin. Pharmacol. 5: 368-74 (2001) and Lonberg, Curr. Opin. Immunol. 20:450-459 (2008).

[0264] Human antibodies may be prepared by administering an immunogen to a transgenic animal that has been modified to produce intact human antibodies or intact antibodies with human variable regions in response to antigenic challenge. Such animals typically contain all or a portion of the human immunoglobulin loci, which replace the endogenous immunoglobulin loci, or which are present extrachromosomally or integrated randomly into the animal's chromosomes. In such transgenic mice, the endogenous immunoglobulin loci

have generally been inactivated. For review of methods for obtaining human antibodies from transgenic animals, see Lonberg, Nat. Biotech. 23:1117-1125 (2005). See also, e.g., U.S. Patent Nos. 6,075,181 and 6,150,584 describing XENOMOUSETM technology; U.S. Patent No. 5,770,429 describing HuMab® technology; U.S. Patent No. 7,041,870 describing K-M MOUSE® technology, and U.S. Patent Application Publication No. US 2007/0061900, describing VelociMouse® technology). Human variable regions from intact antibodies generated by such animals may be further modified, e.g., by combining with a different human constant region.

[0265] Human antibodies can also be made by hybridoma-based methods. Human myeloma and mouse-human heteromyeloma cell lines for the production of human monoclonal antibodies have been described. (See, e.g., Kozbor J. Immunol., 133: 3001 (1984); Brodeur et al., Monoclonal Antibody Production Techniques and Applications, pp. 51-63 (Marcel Dekker, Inc., New York, 1987); and Boerner et al., J. Immunol., 147: 86 (1991).) Human antibodies generated via human B-cell hybridoma technology are also described in Li et al., Proc. Natl. Acad. Sci. USA, 103:3557-3562 (2006). Additional methods include those described, for example, in U.S. Patent No. 7,189,826 (describing production of monoclonal human IgM antibodies from hybridoma cell lines) and Ni, Xiandai Mianyixue, 26(4):265-268 (2006) (describing human-human hybridomas). Human hybridoma technology (Trioma technology) is also described in Vollmers and Brandlein, Histology and Histopathology, 20(3):927-937 (2005) and Vollmers and Brandlein, Methods and Findings in Experimental and Clinical Pharmacology, 27(3):185-91 (2005).

[0266] Human antibodies may also be generated by isolating Fv clone variable domain sequences selected from human-derived phage display libraries. Such variable domain sequences may then be combined with a desired human constant domain. Techniques for selecting human antibodies from antibody libraries are described below.

5. Library-Derived Antibodies

[0267] Antibodies of the invention may be isolated by screening combinatorial libraries for antibodies with the desired activity or activities. For example, a variety of methods are known in the art for generating phage display libraries and screening such libraries for antibodies possessing the desired binding characteristics. Such methods are reviewed, e.g., in Hoogenboom et al. in Methods in Molecular Biology 178:1-37 (O'Brien et al., ed., Human Press, Totowa, NJ, 2001) and further described, e.g., in the McCafferty et al., Nature 348:552-554; Clackson et al., Nature 352: 624-628 (1991); Marks et al., J. Mol. Biol. 222:

581-597 (1992); Marks and Bradbury, in Methods in Molecular Biology 248:161-175 (Lo, ed., Human Press, Totowa, NJ, 2003); Sidhu et al., J. Mol. Biol. 338(2): 299-310 (2004); Lee et al., J. Mol. Biol. 340(5): 1073-1093 (2004); Fellouse, Proc. Natl. Acad. Sci. USA 101(34): 12467-12472 (2004); and Lee et al., J. Immunol. Methods 284(1-2): 119-132(2004).

[0268] In certain phage display methods, repertoires of VH and VL genes are separately cloned by polymerase chain reaction (PCR) and recombined randomly in phage libraries, which can then be screened for antigen-binding phage as described in Winter et al., Ann. Rev. Immunol., 12: 433-455 (1994). Phage typically display antibody fragments, either as single-chain Fv (scFv) fragments or as Fab fragments. Libraries from immunized sources provide high-affinity antibodies to the immunogen without the requirement of constructing hybridomas. Alternatively, the naive repertoire can be cloned (e.g., from human) to provide a single source of antibodies to a wide range of non-self and also self antigens without any immunization as described by Griffiths et al., EMBO J, 12: 725-734 (1993). Finally, naive libraries can also be made synthetically by cloning unrearranged V-gene segments from stem cells, and using PCR primers containing random sequence to encode the highly variable CDR3 regions and to accomplish rearrangement in vitro, as described by Hoogenboom and Winter, J. Mol. Biol., 227: 381-388 (1992). Patent publications describing human antibody phage libraries include, for example: US Patent No. 5,750,373, and US Patent Publication Nos. 2005/0079574, 2005/0119455, 2005/0266000, 2007/0117126, 2007/0160598, 2007/0237764, 2007/0292936, and 2009/0002360.

[0269] Antibodies or antibody fragments isolated from human antibody libraries are considered human antibodies or human antibody fragments herein.

6. Multispecific Antibodies

[0270] In certain embodiments, an antibody provided herein is a multispecific antibody, e.g. a bispecific antibody. Multispecific antibodies are monoclonal antibodies that have binding specificities for at least two different sites. In certain embodiments, one of the binding specificities is for OX40 and the other is for any other antigen. In certain embodiments, bispecific antibodies may bind to two different epitopes of OX40. Bispecific antibodies may also be used to localize cytotoxic agents to cells which express OX40. Bispecific antibodies can be prepared as full length antibodies or antibody fragments.

[0271] Techniques for making multispecific antibodies include, but are not limited to, recombinant co-expression of two immunoglobulin heavy chain-light chain pairs having different specificities (see Milstein and Cuello, Nature 305: 537 (1983)), WO 93/08829, and

Traunecker et al., EMBO J. 10: 3655 (1991)), and "knob-in-hole" engineering (see, e.g., U.S. Patent No. 5,731,168). Multi-specific antibodies may also be made by engineering electrostatic steering effects for making antibody Fc-heterodimeric molecules (WO 2009/089004A1); cross-linking two or more antibodies or fragments (see, e.g., US Patent No. 4,676,980, and Brennan et al., Science, 229: 81 (1985)); using leucine zippers to produce bi-specific antibodies (see, e.g., Kostelny et al., J. Immunol., 148(5):1547-1553 (1992)); using "diabody" technology for making bispecific antibody fragments (see, e.g., Hollinger et al., Proc. Natl. Acad. Sci. USA, 90:6444-6448 (1993)); and using single-chain Fv (sFv) dimers (see,e.g. Gruber et al., J. Immunol., 152:5368 (1994)); and preparing trispecific antibodies as described, e.g., in Tutt et al. J. Immunol. 147: 60 (1991).

[0272] Engineered antibodies with three or more functional antigen binding sites, including "Octopus antibodies," are also included herein (see, e.g. US 2006/0025576A1).

[0273] The antibody or fragment herein also includes a "Dual Acting FAb" or "DAF" comprising an antigen binding site that binds to OX40 as well as another, different antigen (see, US 2008/0069820, for example).

7. Antibody Variants

[0274] In certain embodiments, amino acid sequence variants of the antibodies provided herein are contemplated. For example, it may be desirable to improve the binding affinity and/or other biological properties of the antibody. Amino acid sequence variants of an antibody may be prepared by introducing appropriate modifications into the nucleotide sequence encoding the antibody, or by peptide synthesis. Such modifications include, for example, deletions from, and/or insertions into and/or substitutions of residues within the amino acid sequences of the antibody. Any combination of deletion, insertion, and substitution can be made to arrive at the final construct, provided that the final construct possesses the desired characteristics, e.g., antigen-binding.

a) Substitution, Insertion, and Deletion Variants

[0275] In certain embodiments, antibody variants having one or more amino acid substitutions are provided. Sites of interest for substitutional mutagenesis include the HVRs and FRs. Conservative substitutions are shown in Table A under the heading of "preferred substitutions." More substantial changes are provided in Table A under the heading of "exemplary substitutions," and as further described below in reference to amino acid side chain classes. Amino acid substitutions may be introduced into an antibody of interest and

the products screened for a desired activity, e.g., retained/improved antigen binding, decreased immunogenicity, or improved ADCC or CDC.

TABLE A

Original Residue	Exemplary Substitutions	Preferred Substitutions
Ala (A)	Val; Leu; Ile	Val
Arg (R)	Lys; Gln; Asn	Lys
Asn (N)	Gln; His; Asp, Lys; Arg	Gln
Asp (D)	Glu; Asn	Glu
Cys (C)	Ser; Ala	Ser
Gln (Q)	Asn; Glu	Asn
Glu (E)	Asp; Gln	Asp
Gly (G)	Ala	Ala
His (H)	Asn; Gln; Lys; Arg	Arg
Ile (I)	Leu; Val; Met; Ala; Phe; Norleucine	Leu
Leu (L)	Norleucine; Ile; Val; Met; Ala; Phe	Ile
Lys (K)	Arg; Gln; Asn	Arg
Met (M)	Leu; Phe; Ile	Leu
Phe (F)	Trp; Leu; Val; Ile; Ala; Tyr	Tyr
Pro (P)	Ala	Ala
Ser (S)	Thr	Thr
Thr (T)	Val; Ser	Ser
Trp (W)	Tyr; Phe	Tyr
Tyr (Y)	Trp; Phe; Thr; Ser	Phe
Val (V)	Ile; Leu; Met; Phe; Ala; Norleucine	Leu

[0276] Amino acids may be grouped according to common side-chain properties:

(1) hydrophobic: Norleucine, Met, Ala, Val, Leu, Ile;

(2) neutral hydrophilic: Cys, Ser, Thr, Asn, Gln;

(3) acidic: Asp, Glu;

(4) basic: His, Lys, Arg;

(5) residues that influence chain orientation: Gly, Pro;

(6) aromatic: Trp, Tyr, Phe.

[0277] Non-conservative substitutions will entail exchanging a member of one of these classes for another class.

[0278] One type of substitutional variant involves substituting one or more hypervariable region residues of a parent antibody (e.g. a humanized or human antibody). Generally, the resulting variant(s) selected for further study will have modifications (e.g., improvements) in certain biological properties (e.g., increased affinity, reduced immunogenicity) relative to the parent antibody and/or will have substantially retained certain biological properties of the parent antibody. An exemplary substitutional variant is an affinity matured antibody, which may be conveniently generated, e.g., using phage display-based affinity maturation techniques such as those described herein. Briefly, one or more HVR residues are mutated and the variant antibodies displayed on phage and screened for a particular biological activity (e.g. binding affinity).

[0279] Alterations (e.g., substitutions) may be made in HVRs, e.g., to improve antibody affinity. Such alterations may be made in HVR "hotspots," i.e., residues encoded by codons that undergo mutation at high frequency during the somatic maturation process (see, e.g., Chowdhury, Methods Mol. Biol. 207:179-196 (2008)), and/or residues that contact antigen, with the resulting variant VH or VL being tested for binding affinity. Affinity maturation by constructing and reselecting from secondary libraries has been described, e.g., in Hoogenboom et al. in Methods in Molecular Biology 178:1-37 (O'Brien et al., ed., Human Press, Totowa, NJ, (2001).) In some embodiments of affinity maturation, diversity is introduced into the variable genes chosen for maturation by any of a variety of methods (e.g., error-prone PCR, chain shuffling, or oligonucleotide-directed mutagenesis). A secondary library is then created. The library is then screened to identify any antibody variants with the desired affinity. Another method to introduce diversity involves HVR-directed approaches, in which several HVR residues (e.g., 4-6 residues at a time) are randomized. HVR residues involved in antigen binding may be specifically identified, e.g., using alanine scanning mutagenesis or modeling. CDR-H3 and CDR-L3 in particular are often targeted.

[0280] In certain embodiments, substitutions, insertions, or deletions may occur within one or more HVRs so long as such alterations do not substantially reduce the ability of the antibody to bind antigen. For example, conservative alterations (e.g., conservative substitutions as provided herein) that do not substantially reduce binding affinity may be made in HVRs. Such alterations may, for example, be outside of antigen contacting residues in the HVRs. In certain embodiments of the variant VH and VL sequences provided above,

each HVR either is unaltered, or contains no more than one, two or three amino acid substitutions.

[0281] A useful method for identification of residues or regions of an antibody that may be targeted for mutagenesis is called "alanine scanning mutagenesis" as described by Cunningham and Wells (1989) Science, 244:1081-1085. In this method, a residue or group of target residues (e.g., charged residues such as arg, asp, his, lys, and glu) are identified and replaced by a neutral or negatively charged amino acid (e.g., alanine or polyalanine) to determine whether the interaction of the antibody with antigen is affected. Further substitutions may be introduced at the amino acid locations demonstrating functional sensitivity to the initial substitutions. Alternatively, or additionally, a crystal structure of an antigen-antibody complex to identify contact points between the antibody and antigen. Such contact residues and neighboring residues may be targeted or eliminated as candidates for substitution. Variants may be screened to determine whether they contain the desired properties.

[0282] Amino acid sequence insertions include amino- and/or carboxyl-terminal fusions ranging in length from one residue to polypeptides containing a hundred or more residues, as well as intrasequence insertions of single or multiple amino acid residues. Examples of terminal insertions include an antibody with an N-terminal methionyl residue. Other insertional variants of the antibody molecule include the fusion to the N- or C-terminus of the antibody to an enzyme (e.g. for ADEPT) or a polypeptide which increases the serum half-life of the antibody.

b) Glycosylation variants

[0283] In certain embodiments, an antibody provided herein is altered to increase or decrease the extent to which the antibody is glycosylated. Addition or deletion of glycosylation sites to an antibody may be conveniently accomplished by altering the amino acid sequence such that one or more glycosylation sites is created or removed.

[0284] Where the antibody comprises an Fc region, the carbohydrate attached thereto may be altered. Native antibodies produced by mammalian cells typically comprise a branched, biantennary oligosaccharide that is generally attached by an N-linkage to Asn297 of the CH2 domain of the Fc region. See, e.g., Wright et al. TIBTECH 15:26-32 (1997). The oligosaccharide may include various carbohydrates, e.g., mannose, N-acetyl glucosamine (GlcNAc), galactose, and sialic acid, as well as a fucose attached to a GlcNAc in the "stem" of the biantennary oligosaccharide structure. In some embodiments, modifications of the

oligosaccharide in an antibody of the invention may be made in order to create antibody variants with certain improved properties.

[0285] In one embodiment, antibody variants are provided having a carbohydrate structure that lacks fucose attached (directly or indirectly) to an Fc region. For example, the amount of fucose in such antibody may be from 1% to 80%, from 1% to 65%, from 5% to 65% or from 20% to 40%. The amount of fucose is determined by calculating the average amount of fucose within the sugar chain at Asn297, relative to the sum of all glycostructures attached to Asn 297 (e. g. complex, hybrid and high mannose structures) as measured by MALDI-TOF mass spectrometry, as described in WO 2008/077546, for example. Asn297 refers to the asparagine residue located at about position 297 in the Fc region (Eu numbering of Fc region residues); however, Asn297 may also be located about ± 3 amino acids upstream or downstream of position 297, i.e., between positions 294 and 300, due to minor sequence variations in antibodies. Such fucosylation variants may have improved ADCC function. See, e.g., US Patent Publication Nos. US 2003/0157108 (Presta, L.); US 2004/0093621 (Kyowa Hakko Kogyo Co., Ltd). Examples of publications related to "defucosylated" or "fucose-deficient" antibody variants include: US 2003/0157108; WO 2000/61739; WO 2001/29246; US 2003/0115614; US 2002/0164328; US 2004/0093621; US 2004/0132140; US 2004/0110704; US 2004/0110282; US 2004/0109865; WO 2003/085119; WO 2003/084570; WO 2005/035586; WO 2005/035778; WO2005/053742; WO2002/031140; Okazaki et al. J. Mol. Biol. 336:1239-1249 (2004); Yamane-Ohnuki et al. Biotech. Bioeng. 87: 614 (2004). Examples of cell lines capable of producing defucosylated antibodies include Lec13 CHO cells deficient in protein fucosylation (Ripka et al. Arch. Biochem. Biophys. 249:533-545 (1986); US Pat Appl No US 2003/0157108 A1, Presta, L; and WO 2004/056312 A1, Adams et al., especially at Example 11), and knockout cell lines, such as alpha-1,6-fucosyltransferase gene, FUT8, knockout CHO cells (see, e.g., Yamane-Ohnuki et al. Biotech. Bioeng. 87: 614 (2004); Kanda, Y. et al., Biotechnol. Bioeng., 94(4):680-688 (2006); and WO2003/085107).

[0286] Antibodies variants are further provided with bisected oligosaccharides, e.g., in which a biantennary oligosaccharide attached to the Fc region of the antibody is bisected by GlcNAc. Such antibody variants may have reduced fucosylation and/or improved ADCC function. Examples of such antibody variants are described, e.g., in WO 2003/011878 (Jean-Mairet et al.); US Patent No. 6,602,684 (Umana et al.); and US 2005/0123546 (Umana et al.). Antibody variants with at least one galactose residue in the oligosaccharide attached to the Fc region are also provided. Such antibody variants may have improved CDC function. Such

antibody variants are described, e.g., in WO 1997/30087 (Patel et al.); WO 1998/58964 (Raju, S.); and WO 1999/22764 (Raju, S.).

c) Fc region variants

[0287] In certain embodiments, one or more amino acid modifications may be introduced into the Fc region of an antibody provided herein, thereby generating an Fc region variant. The Fc region variant may comprise a human Fc region sequence (e.g., a human IgG1, IgG2, IgG3 or IgG4 Fc region) comprising an amino acid modification (e.g. a substitution) at one or more amino acid positions.

[0288] In certain embodiments, the invention contemplates an antibody variant that possesses some but not all effector functions, which make it a desirable candidate for applications in which the half life of the antibody in vivo is important yet certain effector functions (such as complement and ADCC) are unnecessary or deleterious. In vitro and/or in vivo cytotoxicity assays can be conducted to confirm the reduction/depletion of CDC and/or ADCC activities. For example, Fc receptor (FcR) binding assays can be conducted to ensure that the antibody lacks FcγR binding (hence likely lacking ADCC activity), but retains FcRn binding ability. The primary cells for mediating ADCC, NK cells, express Fc(RIII only, whereas monocytes express Fc(RI, Fc(RII and Fc(RIII. FcR expression on hematopoietic cells is summarized in Table 3 on page 464 of Ravetch and Kinet, Annu. Rev. Immunol. 9:457-492 (1991). Non-limiting examples of in vitro assays to assess ADCC activity of a molecule of interest is described in U.S. Patent No. 5,500,362 (see, e.g. Hellstrom, I. et al. Proc. Nat'l Acad. Sci. USA 83:7059-7063 (1986)) and Hellstrom, I et al., Proc. Nat'l Acad. Sci. USA 82:1499-1502 (1985); 5,821,337 (see Bruggemann, M. et al., J. Exp. Med. 166:1351-1361 (1987)). Alternatively, non-radioactive assays methods may be employed (see, for example, ACTI™ non-radioactive cytotoxicity assay for flow cytometry (CellTechnology, Inc. Mountain View, CA; and CytoTox 96® non-radioactive cytotoxicity assay (Promega, Madison, WI). Useful effector cells for such assays include peripheral blood mononuclear cells (PBMC) and Natural Killer (NK) cells. Alternatively, or additionally, ADCC activity of the molecule of interest may be assessed in vivo, e.g., in an animal model such as that disclosed in Clynes et al. Proc. Nat'l Acad. Sci. USA 95:652-656 (1998). C1q binding assays may also be carried out to confirm that the antibody is unable to bind C1q and hence lacks CDC activity. See, e.g., C1q and C3c binding ELISA in WO 2006/029879 and WO 2005/100402. To assess complement activation, a CDC assay may be performed (see, for example, Gazzano-Santoro et al., J. Immunol. Methods 202:163 (1996); Cragg, M.S. et

al., Blood 101:1045-1052 (2003); and Cragg, M.S. and M.J. Glennie, Blood 103:2738-2743 (2004)). FcRn binding and in vivo clearance/half life determinations can also be performed using methods known in the art (see, e.g., Petkova, S.B. et al., Int'l. Immunol. 18(12):1759-1769 (2006)).

[0289] In some embodiments, an antibody includes an Fc region with a mutation that decreases binding to an Fc receptor. Antibodies with reduced effector function include without limitation those with substitution of one or more of Fc region residues 238, 265, 269, 270, 297, 327 and 329 (U.S. Patent No. 6,737,056). Such Fc mutants include Fc mutants with substitutions at two or more of amino acid positions 265, 269, 270, 297 and 327, including the so-called "DANA" Fc mutant with substitution of residues 265 and 297 to alanine (US Patent No. 7,332,581).

[0290] Certain antibody variants with improved or diminished binding to FcRs are described. (See, e.g., U.S. Patent No. 6,737,056; WO 2004/056312, and Shields et al., J. Biol. Chem. 9(2): 6591-6604 (2001).)

[0291] In certain embodiments, an antibody variant comprises an Fc region with one or more amino acid substitutions which improve ADCC, e.g., substitutions at positions 298, 333, and/or 334 of the Fc region (EU numbering of residues).

[0292] In some embodiments, alterations are made in the Fc region that result in altered (i.e., either improved or diminished) C1q binding and/or Complement Dependent Cytotoxicity (CDC), e.g., as described in US Patent No. 6,194,551, WO 99/51642, and Idusogie et al. J. Immunol. 164: 4178-4184 (2000).

[0293] Antibodies with increased half lives and improved binding to the neonatal Fc receptor (FcRn), which is responsible for the transfer of maternal IgGs to the fetus (Guyer et al., J. Immunol. 117:587 (1976) and Kim et al., J. Immunol. 24:249 (1994)), are described in US2005/0014934A1 (Hinton et al.). Those antibodies comprise an Fc region with one or more substitutions therein which improve binding of the Fc region to FcRn. Such Fc variants include those with substitutions at one or more of Fc region residues: 238, 256, 265, 272, 286, 303, 305, 307, 311, 312, 317, 340, 356, 360, 362, 376, 378, 380, 382, 413, 424 or 434, e.g., substitution of Fc region residue 434 (US Patent No. 7,371,826).

[0294] See also Duncan & Winter, Nature 322:738-40 (1988); U.S. Patent No. 5,648,260; U.S. Patent No. 5,624,821; and WO 94/29351 concerning other examples of Fc region variants.

d) Cysteine engineered antibody variants

[0295] In certain embodiments, it may be desirable to create cysteine engineered antibodies, e.g., "thioMAbs," in which one or more residues of an antibody are substituted with cysteine residues. In particular embodiments, the substituted residues occur at accessible sites of the antibody. By substituting those residues with cysteine, reactive thiol groups are thereby positioned at accessible sites of the antibody and may be used to conjugate the antibody to other moieties, such as drug moieties or linker-drug moieties, to create an immunoconjugate, as described further herein. In certain embodiments, any one or more of the following residues may be substituted with cysteine: V205 (Kabat numbering) of the light chain; A118 (EU numbering) of the heavy chain; and S400 (EU numbering) of the heavy chain Fc region. Cysteine engineered antibodies may be generated as described, e.g., in U.S. Patent No. 7,521,541.

e) Antibody Derivatives

[0296] In certain embodiments, an antibody provided herein may be further modified to contain additional nonproteinaceous moieties that are known in the art and readily available. The moieties suitable for derivatization of the antibody include but are not limited to water soluble polymers. Non-limiting examples of water soluble polymers include, but are not limited to, polyethylene glycol (PEG), copolymers of ethylene glycol/propylene glycol, carboxymethylcellulose, dextran, polyvinyl alcohol, polyvinyl pyrrolidone, poly-1, 3-dioxolane, poly-1,3,6-trioxane, ethylene/maleic anhydride copolymer, polyaminoacids (either homopolymers or random copolymers), and dextran or poly(n-vinyl pyrrolidone)polyethylene glycol, propropylene glycol homopolymers, prolypropylene oxide/ethylene oxide co-polymers, polyoxyethylated polyols (e.g., glycerol), polyvinyl alcohol, and mixtures thereof. Polyethylene glycol propionaldehyde may have advantages in manufacturing due to its stability in water. The polymer may be of any molecular weight, and may be branched or unbranched. The number of polymers attached to the antibody may vary, and if more than one polymer are attached, they can be the same or different molecules. In general, the number and/or type of polymers used for derivatization can be determined based on considerations including, but not limited to, the particular properties or functions of the antibody to be improved, whether the antibody derivative will be used in a therapy under defined conditions, etc.

[0297] In another embodiment, conjugates of an antibody and nonproteinaceous moiety that may be selectively heated by exposure to radiation are provided. In one embodiment, the nonproteinaceous moiety is a carbon nanotube (Kam et al., Proc. Natl. Acad. Sci. USA 102:

11600-11605 (2005)). The radiation may be of any wavelength, and includes, but is not limited to, wavelengths that do not harm ordinary cells, but which heat the nonproteinaceous moiety to a temperature at which cells proximal to the antibody-nonproteinaceous moiety are killed.

Recombinant Methods and Compositions

[0298] Antibodies may be produced using recombinant methods and compositions, e.g., as described in U.S. Patent No. 4,816,567. In one embodiment, isolated nucleic acid encoding an anti-OX40 antibody described herein is provided. Such nucleic acid may encode an amino acid sequence comprising the VL and/or an amino acid sequence comprising the VH of the antibody (e.g., the light and/or heavy chains of the antibody). In a further embodiment, one or more vectors (e.g., expression vectors) comprising such nucleic acid are provided. In a further embodiment, a host cell comprising such nucleic acid is provided. In one such embodiment, a host cell comprises (e.g., has been transformed with): (1) a vector comprising a nucleic acid that encodes an amino acid sequence comprising the VL of the antibody and an amino acid sequence comprising the VH of the antibody, or (2) a first vector comprising a nucleic acid that encodes an amino acid sequence comprising the VL of the antibody and a second vector comprising a nucleic acid that encodes an amino acid sequence comprising the VH of the antibody. In one embodiment, the host cell is eukaryotic, e.g. a Chinese Hamster Ovary (CHO) cell or lymphoid cell (e.g., Y0, NS0, Sp20 cell). In one embodiment, a method of making an anti-OX40 antibody is provided, wherein the method comprises culturing a host cell comprising a nucleic acid encoding the antibody, as provided above, under conditions suitable for expression of the antibody, and optionally recovering the antibody from the host cell (or host cell culture medium).

[0299] For recombinant production of an anti-OX40 antibody, nucleic acid encoding an antibody, e.g., as described above, is isolated and inserted into one or more vectors for further cloning and/or expression in a host cell. Such nucleic acid may be readily isolated and sequenced using conventional procedures (e.g., by using oligonucleotide probes that are capable of binding specifically to genes encoding the heavy and light chains of the antibody).

[0300] Suitable host cells for cloning or expression of antibody-encoding vectors include prokaryotic or eukaryotic cells described herein. For example, antibodies may be produced in bacteria, in particular when glycosylation and Fc effector function are not needed. For expression of antibody fragments and polypeptides in bacteria, see, e.g., U.S. Patent Nos. 5,648,237, 5,789,199, and 5,840,523. (See also Charlton, Methods in Molecular Biology,

Vol. 248 (B.K.C. Lo, ed., Humana Press, Totowa, NJ, 2003), pp. 245-254, describing expression of antibody fragments in E. coli.) After expression, the antibody may be isolated from the bacterial cell paste in a soluble fraction and can be further purified.

[0301] In addition to prokaryotes, eukaryotic microbes such as filamentous fungi or yeast are suitable cloning or expression hosts for antibody-encoding vectors, including fungi and yeast strains whose glycosylation pathways have been "humanized," resulting in the production of an antibody with a partially or fully human glycosylation pattern. See Gerngross, Nat. Biotech. 22:1409-1414 (2004), and Li et al., Nat. Biotech. 24:210-215 (2006).

[0302] Suitable host cells for the expression of glycosylated antibody are also derived from multicellular organisms (invertebrates and vertebrates). Examples of invertebrate cells include plant and insect cells. Numerous baculoviral strains have been identified which may be used in conjunction with insect cells, particularly for transfection of Spodoptera frugiperda cells.

[0303] Plant cell cultures can also be utilized as hosts. See, e.g., US Patent Nos. 5,959,177, 6,040,498, 6,420,548, 7,125,978, and 6,417,429 (describing PLANTIBODIESTM technology for producing antibodies in transgenic plants).

[0304] Vertebrate cells may also be used as hosts. For example, mammalian cell lines that are adapted to grow in suspension may be useful. Other examples of useful mammalian host cell lines are monkey kidney CV1 line transformed by SV40 (COS-7); human embryonic kidney line (293 or 293 cells as described, e.g., in Graham et al., J. Gen Virol. 36:59 (1977)); baby hamster kidney cells (BHK); mouse sertoli cells (TM4 cells as described, e.g., in Mather, Biol. Reprod. 23:243-251 (1980)); monkey kidney cells (CV1); African green monkey kidney cells (VERO-76); human cervical carcinoma cells (HELA); canine kidney cells (MDCK; buffalo rat liver cells (BRL 3A); human lung cells (W138); human liver cells (Hep G2); mouse mammary tumor (MMT 060562); TRI cells, as described, e.g., in Mather et al., Annals N.Y. Acad. Sci. 383:44-68 (1982); MRC 5 cells; and FS4 cells. Other useful mammalian host cell lines include Chinese hamster ovary (CHO) cells, including DHFR-CHO cells (Urlaub et al., Proc. Natl. Acad. Sci. USA 77:4216 (1980)); and myeloma cell lines such as Y0, NS0 and Sp2/0. For a review of certain mammalian host cell lines suitable for antibody production, see, e.g., Yazaki and Wu, Methods in Molecular Biology, Vol. 248 (B.K.C. Lo, ed., Humana Press, Totowa, NJ), pp. 255-268 (2003).

Assays

[0305] Anti-OX40 antibodies provided herein may be identified, screened for, or characterized for their physical/chemical properties and/or biological activities by various assays known in the art.

1. Binding assays and other assays

[0306] In one aspect, an antibody of the invention is tested for its antigen binding activity, e.g., by known methods such as ELISA, Western blot, etc. OX40 binding may be determined using methods known in the art and exemplary methods are disclosed herein. In one embodiment, binding is measured using radioimmunoassay. An exemplary radioimmunassay is exemplified in the Examples. OX40 antibody is iodinated, and competition reaction mixtures are prepared containing a fixed concentration of iodinated antibody and decreasing concentrations of serially diluted, unlabeled OZ X40 antibody. Cells expressing OX40 (e.g., BT474 cells stably transfected with human OX40) are added to the reaction mixture. Following an incubation, cells are washed to separate the free iodinated OX40 antibody from the OX40 antibody bound to the cells. Level of bound iodinated OX40 antibody is determined, e.g., by counting radioactivity associated with cells, and binding affinity determined using standard methods. In another embodiment, ability of OX40 antibody to bind to surface-expressed OX40 (e.g., on T cell subsets) is assessed using flow cytometry. Peripheral white blood cells are obtained (e.g., from human, cynomolgus monkey, rat or mouse) and cells are blocked with serum. Labeled OX40 antibody is added in serial dilutions, and T cells are also stained to identify T cell subsets (using methods known in the art). Following incubation of the samples and washing, the cells are sorted using flow cytometer, and data analyzed using methods well known in the art. In another embodiment, OX40 binding may be analyzed using surface plasmon resonance. An exemplary surface plasmon resonance method is exemplified in the Examples.

[0307] In another aspect, competition assays may be used to identify an antibody that competes with any of the anti-OX40 antibodies disclosed herein for binding to OX40. In certain embodiments, such a competing antibody binds to the same epitope (e.g., a linear or a conformational epitope) that is bound by any of the anti-OX40 antibodies disclosed herein. Detailed exemplary methods for mapping an epitope to which an antibody binds are provided in Morris (1996) "Epitope Mapping Protocols," in Methods in Molecular Biology vol. 66 (Humana Press, Totowa, NJ). A competition assay is exemplified in the Examples.

[0308] In an exemplary competition assay, immobilized OX40 is incubated in a solution comprising a first labeled antibody that binds to OX40 (e.g., mab 1A7.gr.1, mab 3C8.gr5)

and a second unlabeled antibody that is being tested for its ability to compete with the first antibody for binding to OX40. The second antibody may be present in a hybridoma supernatant. As a control, immobilized OX40 is incubated in a solution comprising the first labeled antibody but not the second unlabeled antibody. After incubation under conditions permissive for binding of the first antibody to OX40, excess unbound antibody is removed, and the amount of label associated with immobilized OX40 is measured. If the amount of label associated with immobilized OX40 is substantially reduced in the test sample relative to the control sample, then that indicates that the second antibody is competing with the first antibody for binding to OX40. See Harlow and Lane (1988) Antibodies: A Laboratory Manual ch.14 (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY).

2. Activity assays

[0309] In one aspect, assays are provided for identifying anti-OX40 antibodies thereof having biological activity. Biological activity may include, e.g., binding OX40 (e.g., binding human and/or cynomolgus OX40), increasing OX40-mediated signal transduction (e.g., increasing NFkB-mediated transcription), depleting cells that express human OX40 (e.g., T cells), depleting cells that express human OX40 by ADCC and/or phagocytosis, enhancing T effector cell function (e.g., CD4+ effector T cell), e.g., by increasing effector T cell proliferation and/or increasing cytokine production (e.g., gamma interferon) by effector T cells, enhancing memory T cell function (e.g., CD4+ memory T cell), e.g., by increasing memory T cell proliferation and/or increasing cytokine production by memory T cells (e.g., gamma interferon), inhibiting regulatory T cell function (e.g., by decreasing Treg suppression of effector T cell function (e.g., CD4+ effector T cell function), binding human effector cells. Antibodies having such biological activity in vivo and/or in vitro are also provided.

[0310] In certain embodiments, an antibody of the invention is tested for such biological activity.

[0311] T cell costimulation may be assayed using methods known in the art and exemplary methods are disclosed herein. For example, T cells (e.g., memory or effector T cells) may be obtained from peripheral white blood cells (e.g., isolated from human whole blood using Ficoll gradient centrifugation). Memory T cells (e.g., CD4+ memory T cells) or effector T cells (e.g. CD4+ Teff cells) may be isolated from PBMC using methods known in the art. For example, the Miltenyi CD4+ memory T cell isolation kit or Miltenyi naïve CD4+ T cell isolation kit may be used. Isolated T cells are cultured in the presence of antigen presenting cells (e.g., irradiated L cells that express CD32 and CD80), and activated by addition of anti-

CD3 antibody in the presence or absence of OX40 agonist antibody. Effect of agonist OX40 antibody of T cell proliferation may be measured using methods well known in the art. For example, the CellTiter Glo kit (Promega) may be used, and results read on a Multilabel Reader (Perkin Elmer). Effect of agonist OX40 antibody on T cell function may also be determined by analysis of cytokines produced by the T cell. In one embodiment, production of interferon gamma by CD4+ T cells is determined, e.g., by measurement of interferon gamma in cell culture supernatant. Methods for measuring interferon gamma are well-known in the art.

[0312] Treg cell function may be assayed using methods known in the art and exemplary methods are disclosed herein. In one example, the ability of Treg to suppress effector T cell proliferation is assayed. T cells are isolated from human whole blood using methods known in the art (e.g., isolating memory T cells or naïve T cells). Purified CD4+ naïve T cells are labeled (e.g., with CFSE) and purified Treg cells are labeled with a different reagent. Irradiated antigen presenting cells (e.g., L cells expressing CD32 and CD80) are co-cultured with the labeled purified naïve CD4+ T cells and purified Tregs. The co-cultures are activated using anti-CD3 antibody and tested in the presence or absence of agonist OX40 antibody. Following a suitable time (e.g., 6 days of coculture), level of CD4+ naïve T cell proliferation is tracked by dye dilution in reduced label staining (e.g., reduced CFSE label staining) using FACS analysis.

[0313] OX40 signaling may be assayed using methods well known in the art and exemplary methods are disclosed herein. In one embodiment, transgenic cells are generated that express human OX40 and a reporter gene comprising the NFkB promoter fused to a reporter gene (e.g., beta luciferase). Addition of OX40 agonist antibody to the cells results in increased NFkB transcription, which is detected using an assay for the reporter gene.

[0314] Phagocytosis may be assayed, e.g., by using monocyte-derived macrophages, or U937 cells (a human histiocytic lymphoma cells line with the morphology and characteristics of mature macrophages). OX40 expressing cells are added to the monocyte-derived macrophages or U937 cells in the presence or absence of anti-OX40 agonist antibody. Following culturing of the cells for a suitable period of time, the percentage of phagocytosis is determined by examining percentage of cells that double stain for markers of 1) the macrophage or U937 cell and 2) the OX40 expressing cell, and dividing this by the total number of cells that show markers of the OX40 expressing cell (e.g., GFP). Analysis may be done by flow cytometry. In another embodiment, analysis may be done by fluorescent microscopy analysis.

[0315] ADCC may be assayed, e.g., using methods well known in the art. Exemplary methods are described in the definition section and an exemplary assay is disclosed in the Examples. In some embodiments, level of OX40 is characterized on an OX40 expressing cell that is used for testing in an ADCC assay. The cell may be stained with a detectably labeled anti-OX40 antibody (e.g., PE labeled), then level of fluorescence determined using flow cytometry, and results presented as median fluorescence intensity (MFI). In another embodiment, ADCC may be analyzed by CellTiter Glo assay kit and cell viability/cytotoxicity may be determined by chemioluminescence.

[0316] The binding affinities of various antibodies to FcγRIA, FcγRIIA, FcγRIIB, and two allotypes of FcγRIIIA (F158 and V158) may be measured in ELISA-based ligand-binding assays using the respective recombinant Fcγ receptors. Purified human Fcγ receptors are expressed as fusion proteins containing the extracellular domain of the receptor γ chain linked to a Gly/6xHis/glutathione S-transferase (GST) polypeptide tag at the C-terminus. The binding affinities of antibodies to those human Fcγ receptors are assayed as follows. For the low-affinity receptors, i.e. FcγRIIA (CD32A), FcγRIIB (CD32B), and the two allotypes of FcγRIIIA (CD16), F-158 and V-158, antibodies may be tested as multimers by cross-linking with a F(ab')2 fragment of goat anti-human kappa chain (ICN Biomedical; Irvine, CA) at an approximate molar ratio of 1:3 antibody:cross-linking F(ab')2. Plates are coated with an anti-GST antibody (Genentech) and blocked with bovine serum albumin (BSA). After washing with phosphate-buffered saline (PBS) containing 0.05% Tween-20 with an ELx405™ plate washer (Biotek Instruments; Winooski, VT), Fcγ receptors are added to the plate at 25 ng/well and incubated at room temperature for 1 hour. After the plates are washed, serial dilutions of test antibodies are added as multimeric complexes and the plates were incubated at room temperature for 2 hours. Following plate washing to remove unbound antibodies, the antibodies bound to the Fcγ receptor are detected with horseradish peroxidase (HRP)–conjugated F(ab')2 fragment of goat anti-human F(ab')2 (Jackson ImmunoResearch Laboratories; West Grove, PA) followed by the addition of substrate, tetramethylbenzidine (TMB) (Kirkegaard & Perry Laboratories; Gaithersburg, MD). The plates are incubated at room temperature for 5–20 minutes, depending on the Fcγ receptors tested, to allow color development. The reaction is terminated with 1 M H3PO4 and absorbance at 450 nm was measured with a microplate reader (SpectraMax®190, Molecular Devices; Sunnyvale, CA). Dose-response binding curves are generated by plotting the mean absorbance values from the duplicates of antibody dilutions against the concentrations of the antibody. Values for the effective concentration of the antibody at which 50% of the maximum response from binding

to the Fcγ receptor is detected (EC50) were determined after fitting the binding curve with a four-parameter equation using SoftMax Pro (Molecular Devices).

[0317] To select for antibodies which induce cell death, loss of membrane integrity as indicated by, e.g., propidium iodide (PI), trypan blue or 7AAD uptake may be assessed relative to control. A PI uptake assay can be performed in the absence of complement and immune effector cells. OX40 expressing cells are incubated with medium alone or medium containing of the appropriate monoclonal antibody at e.g., about 10μg/ml. The cells are incubated for a time period (e.g., 1 or 3 days). Following each treatment, cells are washed and aliquoted. In some embodiments, cells are aliquoted into 35 mm strainer-capped 12 x 75 tubes (1ml per tube, 3 tubes per treatment group) for removal of cell clumps. Tubes then receive PI (10μg/ml). Samples may be analyzed using a FACSCAN™ flow cytometer and FACSCONVERT™ CellQuest software (Becton Dickinson).

[0318] Cells for use in any of the above in vitro assays include cells or cell lines that naturally express OX40 or that have been engineered to express OX40. Such cells include activated T cells, Treg cells and activated memory T cells that naturally express OX40. Such cells also include cell lines that express OX40 and cell lines that do not normally express OX40 but have been transfected with nucleic acid encoding OX40. Exemplary cell lines provided herein for use in any of the above in vitro assays include transgenic BT474 cells (a human breast cancer cell line) that express human OX40

[0319] It is understood that any of the above assays may be carried out using an immunoconjugate of the invention in place of or in addition to an anti-OX40 antibody.

[0320] It is understood that any of the above assays may be carried out using anti-OX40 antibody and an additional therapeutic agent.

Immunoconjugates

[0321] The invention also provides immunoconjugates comprising an anti-OX40 antibody herein conjugated to one or more cytotoxic agents, such as chemotherapeutic agents or drugs, growth inhibitory agents, toxins (e.g., protein toxins, enzymatically active toxins of bacterial, fungal, plant, or animal origin, or fragments thereof), or radioactive isotopes.

[0322] In one embodiment, an immunoconjugate is an antibody-drug conjugate (ADC) in which an antibody is conjugated to one or more drugs, including but not limited to a maytansinoid (see U.S. Patent Nos. 5,208,020, 5,416,064 and European Patent EP 0 425 235 B1); an auristatin such as monomethylauristatin drug moieties DE and DF (MMAE and MMAF) (see U.S. Patent Nos. 5,635,483 and 5,780,588, and 7,498,298); a dolastatin; a

calicheamicin or derivative thereof (see U.S. Patent Nos. 5,712,374, 5,714,586, 5,739,116, 5,767,285, 5,770,701, 5,770,710, 5,773,001, and 5,877,296; Hinman et al., Cancer Res. 53:3336-3342 (1993); and Lode et al., Cancer Res. 58:2925-2928 (1998)); an anthracycline such as daunomycin or doxorubicin (see Kratz et al., Current Med. Chem. 13:477-523 (2006); Jeffrey et al., Bioorganic & Med. Chem. Letters 16:358-362 (2006); Torgov et al., Bioconj. Chem. 16:717-721 (2005); Nagy et al., Proc. Natl. Acad. Sci. USA 97:829-834 (2000); Dubowchik et al., Bioorg. & Med. Chem. Letters 12:1529-1532 (2002); King et al., J. Med. Chem. 45:4336-4343 (2002); and U.S. Patent No. 6,630,579); methotrexate; vindesine; a taxane such as docetaxel, paclitaxel, larotaxel, tesetaxel, and ortataxel; a trichothecene; and CC1065.

[0323] In another embodiment, an immunoconjugate comprises an antibody as described herein conjugated to an enzymatically active toxin or fragment thereof, including but not limited to diphtheria A chain, nonbinding active fragments of diphtheria toxin, exotoxin A chain (from Pseudomonas aeruginosa), ricin A chain, abrin A chain, modeccin A chain, alpha-sarcin, Aleurites fordii proteins, dianthin proteins, Phytolaca americana proteins (PAPI, PAPII, and PAP-S), momordica charantia inhibitor, curcin, crotin, sapaonaria officinalis inhibitor, gelonin, mitogellin, restrictocin, phenomycin, enomycin, and the tricothecenes.

[0324] In another embodiment, an immunoconjugate comprises an antibody as described herein conjugated to a radioactive atom to form a radioconjugate. A variety of radioactive isotopes are available for the production of radioconjugates. Examples include At211, I131, I125, Y90, Re186, Re188, Sm153, Bi212, P32, Pb212 and radioactive isotopes of Lu. When the radioconjugate is used for detection, it may comprise a radioactive atom for scintigraphic studies, for example tc99m or I123, or a spin label for nuclear magnetic resonance (NMR) imaging (also known as magnetic resonance imaging, mri), such as iodine-123 again, iodine-131, indium-111, fluorine-19, carbon-13, nitrogen-15, oxygen-17, gadolinium, manganese or iron.

[0325] Conjugates of an antibody and cytotoxic agent may be made using a variety of bifunctional protein coupling agents such as N-succinimidyl-3-(2-pyridyldithio) propionate (SPDP), succinimidyl-4-(N-maleimidomethyl) cyclohexane-1-carboxylate (SMCC), iminothiolane (IT), bifunctional derivatives of imidoesters (such as dimethyl adipimidate HCl), active esters (such as disuccinimidyl suberate), aldehydes (such as glutaraldehyde), bis-azido compounds (such as bis (p-azidobenzoyl) hexanediamine), bis-diazonium derivatives (such as bis-(p-diazoniumbenzoyl)-ethylenediamine), diisocyanates (such as toluene 2,6-diisocyanate), and bis-active fluorine compounds (such as 1,5-difluoro-2,4-dinitrobenzene).

For example, a ricin immunotoxin can be prepared as described in Vitetta et al., Science 238:1098 (1987). Carbon-14-labeled 1-isothiocyanatobenzyl-3-methyldiethylene triaminepentaacetic acid (MX-DTPA) is an exemplary chelating agent for conjugation of radionucleotide to the antibody. See WO94/11026. The linker may be a "cleavable linker" facilitating release of a cytotoxic drug in the cell. For example, an acid-labile linker, peptidase-sensitive linker, photolabile linker, dimethyl linker or disulfide-containing linker (Chari et al., Cancer Res. 52:127-131 (1992); U.S. Patent No. 5,208,020) may be used.

[0326] The immunuoconjugates or ADCs herein expressly contemplate, but are not limited to such conjugates prepared with cross-linker reagents including, but not limited to, BMPS, EMCS, GMBS, HBVS, LC-SMCC, MBS, MPBH, SBAP, SIA, SIAB, SMCC, SMPB, SMPH, sulfo-EMCS, sulfo-GMBS, sulfo-KMUS, sulfo-MBS, sulfo-SIAB, sulfo-SMCC, and sulfo-SMPB, and SVSB (succinimidyl-(4-vinylsulfone)benzoate) which are commercially available (e.g., from Pierce Biotechnology, Inc., Rockford, IL., U.S.A).

IV. Methods for Predicting Responsiveness

[0327] Provided herein are methods for predicting responsiveness of a subject having cancer to an OX40 agonist treatment and methods for identifying or selecting a subject having cancer for treating with an OX40 agonist. In some embodiments, the methods include measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the one or more marker genes are selected from CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and classifying the subject as a responsive or non-responsive subject based on the expression level of the one or more marker genes in the sample obtained from the subject, as compared with a reference, where an increased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment, or where a decreased expression level of the one or more marker genes as compared with the reference indicates the subject may not be responsive to an OX40 agonist treatment. In some embodiments, the one or more marker genes may be selected from CD8a, CD8b, IFNg, GZMA, GZMB, PRF1, and PDCA1. In some embodiments, the one or more marker genes may be selected from H2-d, CTLA4, CXCL9, PTPRC, IL7R, KLRK1, and CXCL1. In some embodiments, the one or more marker genes may be selected from CD64, IDO1, and ITGAM.

[0328] In other embodiments, the methods include measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the one or more marker genes are selected from CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and classifying the subject as a responsive or non-responsive subject based on the expression level of the one or more marker genes in the sample obtained from the subject, as compared with a reference, wherein a decreased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment, or wherein an increased expression level of the one or more marker genes as compared with the reference indicates the subject may not be responsive to an OX40 agonist treatment.

[0329] Certain aspects of the present disclosure relate to measuring the expression level of one or more marker genes. Any suitable method for measuring gene expression known in the art may be used. In some embodiments, expression level may refer to mRNA expression level. mRNA expression level may be measured by many methods. Such methods may quantify the copies of a specific mRNA present in a sample by measuring the amount of hybridization to an mRNA-specific probe. Other methods may amplify mRNA, or cDNA generated from mRNA, and quantify the amount of amplicon generated to extrapolate how much mRNA was present in a sample. Yet other methods may involve next-generation sequencing of part or all of mRNA transcripts, or cDNA generated from mRNA, then quantifying the number of sequences detected that correspond to particular gene(s). In some embodiments, mRNA expression level is measured by quantitative PCR, semi-quantitative PCR, nucleotide microarray, RNA-seq, in situ hybridization, and/or Northern blotting.

[0330] In some embodiments, expression level may refer to protein expression level. Protein expression level may be measured by many methods. Such methods may quantify proteins present in a sample by using a probe that specifically binds to a particular protein, such as an antibody, then detecting the amount of specific binding in a sample. Other methods may fragment proteins into short peptides, then detect these peptides and quantify how many peptides correspond to particular protein(s). In some embodiments, protein expression level is measured by Western blotting, peptide microarray, immunohistochemistry, flow cytometry, and/or mass spectrometry.

[0331] As described herein, some marker genes may be listed by the name of a murine homolog. In some embodiments, the expression level of a human homolog of one or more marker genes described herein may be measured in a human sample. Methods for determining a human homolog of a murine gene are known in the art. In some embodiments,

a homolog may be functionally determined, i.e., by performing a similar cellular function. In some embodiments, a homolog may be determined by sequence homology, e.g., by using a program such as BLAST, BLAST-2, ALIGN, Megalign (DNASTAR), or ALIGN-2 software as described herein.

[0332] In some embodiments, the marker gene may be one or more of the marker genes provided in Table 3 below. Table 3 provides a list of genes whose expression level was discovered as described herein to be useful for predicting responsiveness to an OX40 agonist treatment. In Table 3, positive predictors are those genes that, when expressed at an increased level, predict responsiveness to an OX40 agonist treatment. Negative predictors are those genes that, when expressed at a decreased level, predict responsiveness to an OX40 agonist treatment. Exemplary gene/protein names or aliases, as well as an exemplary accession number corresponding to a human protein homolog, are also provided.

Table 3.

NO.	Gene	Predictor	Alias (non-exhaustive)	Accession No
1	CSF2	Negative	Granulocyte-macrophage colony-stimulating factor; GM-CSF; CSF;	NM_000749
2	CCL22	Negative	Chemokine (C-C motif) ligand 22; CC chemokine STCP-1; small inducible cytokine A22	NM_022990
3	GATA3	Negative	GATA-binding factor 3, trans-acting T-cell-specific transcription factor GATA3	NM_001002295
4	CD8a	Positive	T-cell surface glycoprotein CD8 alpha chain; OKT8 T-cell antigen; T-cell antigen Leu2; CD8 antigen, alpha polypeptide p32	NM_001145873
5	CD8b	Positive	T-cell surface glycoprotein CD8 beta chain; CD8 antigen, alpha polypeptide p37	NM_172213
6	H2-d	Positive	MHC Class I B	NM_005514
7	CTLA4	Positive	Cytotoxic T-lymphocyte protein 4; CD152 isoform; celiac disease 3; insulin-dependent diabetes mellitus 12	NM_005214
8	CD64	Positive	Fc-gamma RI; Fc-gamma RIA	NM_000566
9	CXCL9	Positive	C-X-C motif chemokine 9; small-inducible cytokine B9; gamma-interferon-induced monokine	NM_002416
10	IFNg	Positive	Interferon gamma	NM_000619
11	IDO1	Positive	Indoleamine 2,3-dioxygenase 1	NM_002164
12	GZMA	Positive	Granzyme A; h factor; CTL tryptase; fragmentin-1; cytotoxic T-lymphocyte proteinase 1; Hanukah factor serine protease	NM_006144

13	GZMB	Positive	Granzyme B; C11; CTLA-1; fragmentin-2; cathepsin G-like 1; T-cell serine protease 1-3E; cytotoxic T-lymphocyte proteinase 2	NM_004131
14	PRF1	Positive	Perforin-1; cytolysin; lymphocyte pore-forming protein	NM_001083116
15	PDCA1	Positive	Bone marrow stromal cell antigen 2; NPC-A-7; HM1.24 antigen	NM_004335
16	KLRK1	Positive	Killer cell lectin-like receptor subfamily K, member 1; NKG2-D type II integral membrane protein; NK cell receptor D	NM_007360
17	PTPRC	Positive	Receptor-type tyrosine-protein phosphatase C; CD45 antigen; T200 glycoprotein	NM_002838
18	CXCL1	Positive	Growth-regulated alpha protein; C-X-C motif chemokine 1; MGSA alpha; fibroblast secretory protein; GRO1 oncogene	NM_001511
19	ITGAM	Positive	Integrin alpha-M; CR-3 alpha chain; antigen CD11b (p170); leukocyte adhesion receptor MO1; neutrophil adherence receptor alpha-M subunit; macrophage antigen alpha polypeptide	NM_001145808
20	IL7R	Positive	Interleukin-7 receptor subunit alpha; CD127 antigen	NM_002185
21	IL13	Negative	Interleukin-13	NM_002188
22	EPCAM	Negative	Epithelial cell adhesion molecule; cell surface glycoprotein Trop-1; adenocarcinoma-associated antigen; GA733-2; epithelial glycoprotein 314	NM_002354
23	VTCN1	Negative	V-set domain-containing T-cell activation inhibitor 1; B7-H4	NM_024626

[0333] In some embodiments, expression level of an mRNA or protein may be normalized to the expression level of a reference gene. Normalizing the expression level of a particular gene to a reference is thought to enhance reproducibility across samples by factoring differences in sample size and/or mRNA/protein extraction. In these examples, expression level relative to the reference is measured. In some embodiments, multiple reference genes may be used, either singly or in aggregate (e.g., by averaging). In other embodiments, expression level of an mRNA or protein may refer to absolute expression level.

[0334] In some embodiments, a reference gene may be a housekeeping gene. A housekeeping gene is thought to be constitutively expressed in a cell in normal and/or pathological states, such as a gene encoding a protein required for basic cellular function and/or maintenance. Housekeeping genes are typically used as a reference to ensure they will be expressed at a detectable and/or reproducible level across multiple samples. Exemplary

housekeeping genes and further description of the use of such genes as a reference may be found, for example, in de Kok, J.B., *et al.* (2005) *Lab Invest.* 85(1):154-9.

[0335] In some embodiments, the expression level of one or more marker genes described herein is compared to a reference. In some embodiments, the reference is the average, mean, or median level of expression of the corresponding marker gene in a sample comprising leukocytes from subjects that have cancer. In some embodiments, the reference is the average, mean, or median level of expression of the corresponding marker gene in samples comprising leukocytes from other subjects having cancer who are not responsive to the OX40 agonist treatment after receiving treatment. For example, a set of samples obtained from cancers having a shared characteristic (e.g., the same cancer type and/or stage, or exposure to a common treatment such as an OX40 agonist) may be studied from a population, such as with a clinical outcome study. This set may be used to derive a reference, e.g., a reference number, to which a subject's sample may be compared.

[0336] In some embodiments, responsiveness to treatment may refer to any one or more of: extending survival (including overall survival and progression free survival); resulting in an objective response (including a complete response or a partial response); or improving signs or symptoms of cancer. In some embodiments, responsiveness may refer to improvement of one or more factors according to the published set of RECIST or Immune-Related Response Criteria guidelines for determining the status of a tumor in a cancer patient, *i.e.,* responding, stabilizing, or progressing. For a more detailed discussion of these guidelines, see Eisenhauer et al., *Eur J Cancer* 2009;45: 228–47; Topalian et al., *N Engl J Med* 2012;366:2443–54; Wolchok et al., *Clin Can Res* 2009;15:7412–20; and Therasse, P., *et al. J. Natl. Cancer Inst.* 92:205-16 (2000). A responsive subject may refer to a subject whose cancer(s) show improvement, e.g., according to one or more factors based on RECIST or Immune-Related Response criteria. A non-responsive subject may refer to a subject whose cancer(s) do not show improvement, e.g., according to one or more factors based on RECIST or Immune-Related Response criteria. In some embodiments, responsiveness may include immune activation. In some embodiments, responsiveness may include treatment efficacy. In some embodiments, responsiveness may include immune activation and treatment efficacy.

[0337] Conventional response criteria may not be adequate to characterize the anti-tumor activity of immunotherapeutic agents, which can produce delayed responses that may be preceded by initial apparent radiological progression, including the appearance of new lesions. Therefore, modified response criteria have been developed that account for the possible appearance of new lesions and allow radiological progression to be confirmed at a subsequent assessment. Accordingly, in

some embodiments, responsiveness may refer to improvement of one of more factors according to immune-related response criteria (irRC). See, e.g., Wolchok et al., Clin Can Res 2009;15:7412 – 20. In some embodiments, new lesions are added into the defined tumor burden and followed, e.g., for radiological progression at a subsequent assessment. In some embodiments, presence of non-target lesions are included in assessment of complete response and not included in assessment of radiological progression. In some embodiments, radiological progression may be determined only on the basis of measurable disease and/or may be confirmed by a consecutive assessment ≥ 4 weeks from the date first documented.

[0338] In some embodiment, tumor may refer to a physical mass containing a plurality of cancer cells, *e.g.,* cells showing the characteristics of any of the cancers described herein. Examples of tumors may include primary tumors of any of the above types of cancer or metastatic tumors at a second site derived from any of the above types of cancer. In some embodiments, a tumor may contain cancer cells as well as tumor stroma.

[0339] Certain aspects of the present disclosure relate to measurement of the expression level of one or more genes in a sample. In some embodiments, a sample may include leukocytes. In some embodiments, the sample may be a tumor sample. A tumor sample may include cancer cells, lymphocytes, leukocytes, stroma, blood vessels, connective tissue, basal lamina, and any other cell type in association with the tumor. In some embodiments, the sample is a tumor tissue sample containing tumor-infiltrating leukocytes. As used herein, any leukocyte associated with a tumor may be considered a tumor-infiltrating leukocyte. Examples of tumor-infiltrating leukocytes include without limitation T lymphocytes (such as CD8+ T lymphocytes and/or CD4+ T lymphocytes), B lymphocytes, or other bone marrow-lineage cells including granulocytes (neutrophils, eosinophils, basophils), monocytes, macrophages, dendritic cells (i.e., interdigitating dendritic cells), histiocytes, and natural killer cells. In some embodiments, a tumor-infiltrating leukocyte may be associated with cancer cells of a tumor. In some embodiments, a tumor-infiltrating leukocyte may be associated with tumor stroma. In some embodiments, the tumor samples are enriched for tumor area by macrodissection.

[0340] In some embodiments, the sample may be processed to separate or isolate one or more cell types (e.g., leukocytes). In some embodiments, the sample may be used without separating or isolating cell types. A tumor sample may be obtained from a subject by any method known in the art, including without limitation a biopsy, endoscopy, or surgical procedure. In some embodiments, a tumor sample may be prepared by methods such as freezing, fixation (e.g., by using formalin or a similar fixative), and/or embedding in paraffin

wax. In some embodiments, a tumor sample may be sectioned. In some embodiments, a fresh tumor sample (i.e., one that has not been prepared by the methods described above) may be used. In some embodiments, a sample may be prepared by incubation in a solution to preserve mRNA and/or protein integrity. A tumor sample containing leukocytes may be assayed by any technique described herein for measuring marker gene expression level.

[0341] In some embodiments, the sample may be a peripheral blood sample. A peripheral blood sample may include white blood cells, PBMCs, and the like. Any technique known in the art for isolating leukocytes from a peripheral blood sample may be used. For example, a blood sample may be drawn, red blood cells may be lysed, and a white blood cell pellet may be isolated and used for the sample. In another example, density gradient separation may be used to separate leukocytes (e.g., PBMCs) from red blood cells. Isolated leukocytes from a peripheral blood sample may be assayed by any technique described herein for measuring marker gene expression level.

V. Methods of Monitoring Pharmacodynamic Activity and Responsiveness

[0342] Provided herein are methods for monitoring pharmacodynamic activity of an OX40 agonist treatment by measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, where the subject has been treated with an OX40 agonist, and where the one or more marker genes are selected from ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1, PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and TNFSF4; and determining the treatment as demonstrating pharmacodynamic activity based on the expression level of the one or more marker genes in the sample obtained from the subject, as compared with a reference, where an increased expression level of the one or more marker genes as compared with the reference indicates pharmacodynamic activity to the OX40 agonist treatment. These methods are based in part on the discovery described herein that expression of specific marker genes (e.g., ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1, PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and/or TNFSF4) is upregulated

following treatment with an OX40 agonist in tumors that are responsive to such treatment and tumors that are non-responsive to such treatment. Expression level of a marker gene may be measured by one or more methods as described herein.

[0343] As used herein, "pharmacodynamic (PD) activity" may refer to an effect of a treatment (e.g., an OX40 agonist treatment) to the subject. An example of a PD activity may include modulation of the expression level of one or more genes. Without wishing to be bound to theory, it is thought that monitoring PD activity, such as by measuring expression of a gene marker, may be advantageous during a clinical trial examining an OX40 agonist. Monitoring PD activity may be used, for example, to monitor response to treatment, toxicity, and the like.

[0344] In some embodiments, the expression level of one or more marker genes may be compared to a reference which may include a sample from a subject not receiving a treatment (e.g., an OX40 agonist treatment). In some embodiments, a reference may include a sample from the same subject before receiving a treatment (e.g., an OX40 agonist treatment). In some embodiments, a reference may include a reference value from one or more samples of other subjects receiving a treatment (e.g., an OX40 agonist treatment). For example, a population of patients may be treated, and a mean, average, or median value for expression level of one or more genes may be generated from the population as a whole. A set of samples obtained from cancers having a shared characteristic (e.g., the same cancer type and/or stage, or exposure to a common treatment such as an OX40 agonist) may be studied from a population, such as with a clinical outcome study. This set may be used to derive a reference, e.g., a reference number, to which a subject's sample may be compared. Any of the references described herein may be used as a reference for monitoring PD activity.

[0345] In some embodiments, the one or more marker genes are selected from CD3, CD8, IFNg, GZMA, GZMB, PRF1, TNFa, PDCD1, and CD274. In some embodiments, the marker gene may be one or more of the marker genes provided in Table 4 below. Table 4 provides a list of genes whose expression level was discovered as described herein to be upregulated in response OX40 agonist treatment (e.g., as a marker of PD activity). Exemplary gene/protein names or aliases, as well as an exemplary accession number corresponding to a human protein homolog, are provided.

Table 4.

NO.	Gene	Alias (non-exhaustive)	Accession No
1	ARG1	Arginase-1; liver-type arginase	NM_001244438

2	CCL2	C-C motif cytokine 2; small-inducible cytokine 2; monocyte chemoattractant protein 1	NM_002982
3	CCL22	Chemokine (C-C motif) ligand 22; CC chemokine STCP-1; small inducible cytokine A22	NM_022990
4	CCL5	C-C motif cytokine 5; beta chemokine RANTES; T-cell specific protein p288; eosinophil chemotactic cytokine	NM_001278736
5	CCR5	C-C chemokine receptor 5; HIV-1 fusion coreceptor	NM_001100168
6	CD226	DNAX accessory molecule-1; platelet and T cell activation antigen 1; T lineage-specific activation antigen 1	NM_006656
7	CD27	CD27L receptor; T cell activation antigen S152; T-cell activation antigen CD27; tumor necrosis factor receptor superfamily, member 7	NM_001242
8	CD274	PD-L1; B7-H1; B7-4; B7-H	NM_014143
9	CD28	T-cell-specific surface glycoprotein CD28	NM_006139
10	CD3E	T-cell surface glycoprotein CD3 epsilon chain; CD3-epsilon; T-cell surface antigen T3/Leu-4 epsilon chain; CD3e antigen, epsilon polypeptide (TiT3 complex); T-cell antigen receptor complex, epsilon subunit of T3	NM_000733
11	CD40	tumor necrosis factor receptor superfamily member 5; CD40L receptor; B cell-associated molecule; nerve growth factor receptor-related B-lymphocyte activation molecule	NM_001250
12	CD8A	T-cell surface glycoprotein CD8 alpha chain; OKT8 T-cell antigen; T-cell antigen Leu2; CD8 antigen, alpha polypeptide p32	NM_001145873
13	CD8b	T-cell surface glycoprotein CD8 beta chain; CD8 antigen, alpha polypeptide p37	NM_172213
14	CXCL10	C-X-C motif chemokine 10; gamma IP10; small-inducible cytokine B10; interferon-inducible cytokine IP-10; 10 kDa interferon gamma-induced protein; small inducible cytokine subfamily B (Cys-X-Cys), member 10	NM_001565
15	CXCL9	C-X-C motif chemokine 9; small-inducible cytokine B9; gamma-interferon-induced monokine	NM_002416
16	EOMES	eomesodermin homolog; T-box brain protein 2	NM_001278182
17	FasL	tumor necrosis factor ligand superfamily member 6; CD95 ligand; fas antigen ligand; apoptosis (APO-1) antigen ligand 1	NM_000639
18	Fcgr1/CD64	Fc-gamma RI; Fc-gamma RIA	NM_000566
19	FOXP3	forkhead box protein P3; scurfin; FOXP3delta7; immunodeficiency, polyendocrinopathy, enteropathy, X-linked	NM_014009
20	GZMA	Granzyme A; h factor; CTL tryptase; fragmentin-1; cytotoxic T-lymphocyte proteinase 1; Hanukah factor serine protease	NM_006144
21	GZMB	Granzyme B; C11; CTLA-1; fragmentin-2; cathepsin G-like 1; T-cell serine protease 1-3E; cytotoxic T-lymphocyte proteinase 2	NM_004131
22	HAVCR2	hepatitis A virus cellular receptor 2; kidney injury molecule-3; T-cell membrane protein 3; T cell immunoglobulin mucin 3; T-cell immunoglobulin	NM_032782

		mucin receptor 3	
23	ICAM1	intercellular adhesion molecule 1; cell surface glycoprotein P3.58; major group rhinovirus receptor	NM_000201
24	IDO1	Indoleamine 2,3-dioxygenase 1	NM_002164
25	IFNg	Interferon gamma	NM_000619
26	IL10	Interleukin-10; T-cell growth inhibitory factor; cytokine synthesis inhibitory factor	NM_000572
27	IL12A (TDO2)	interleukin-12 subunit alpha; CLMF p35; IL-12, subunit p35; interleukin-12 alpha chain; NF cell stimulatory factor chain 1; NK cell stimulatory factor chain 1; cytotoxic lymphocyte maturation factor 1, p35	NM_000882
28	IL13	Interleukin-13	NM_002188
29	IL2	interleukin-2; aldesleukin; T cell growth factor	NM_000586
30	IL7R	Interleukin-7 receptor subunit alpha; CD127 antigen	NM_002185
31	ITGAM	Integrin alpha-M; CR-3 alpha chain; antigen CD11b (p170); leukocyte adhesion receptor MO1; neutrophil adherence receptor alpha-M subunit; macrophage antigen alpha polypeptide	NM_001145808
32	KLRK1	Killer cell lectin-like receptor subfamily K, member 1; NKG2-D type II integral membrane protein; NK cell receptor D	NM_007360
33	LAG3	lymphocyte activation gene 3 protein	NM_002286
34	MAP4K1	mitogen-activated protein kinase kinase kinase kinase 1; MEKKK 1; MEK kinase kinase 1; MAPK/ERK kinase kinase kinase 1; hematopoietic progenitor kinase 1	NM_001042600
35	MS4A1	B-lymphocyte antigen CD20; leukocyte surface antigen Leu-16; B-lymphocyte cell-surface antigen B1	NM_021950
36	PDCD1	CD279; SLEB2	NM_005018
37	PDCD1LG2	B7-DC; Btdc; CD273; PD-L2	NM_025239
38	PRF1	Perforin-1; cytolysin; lymphocyte pore-forming protein	NM_001083116
39	PTPRC	Receptor-type tyrosine-protein phosphatase C; CD45 antigen; T200 glycoprotein	NM_002838
40	TNF	tumor necrosis factor; TNF-a; cachectin; APC1 protein; TNF, monocyte- macrophage-derived; tumor necrosis factor ligand superfamily member 2	NM_000594
41	TNFRSF14	tumor necrosis factor receptor superfamily member 14; CD40-like protein; herpes virus entry mediator A; tumor necrosis factor receptor-like gene2	NM_003820
42	TNFRSF9	tumor necrosis factor receptor superfamily member 9; CD137 antigen; T cell antigen ILA; 4-1BB ligand receptor; induced by lymphocyte activation (ILA); interleukin-activated receptor, homolog of mouse Ly63	NM_001561
43	TNFSF4	tumor necrosis factor ligand superfamily member 4; CD134 ligand; glycoprotein Gp34; OX40 antigen ligand; tax-transcriptionally activated glycoprotein 1 (34kD)	NM_003326

[0346] Provided herein are methods for monitoring responsiveness of a subject to an OX40 agonist treatment by measuring the expression level of one or more marker genes in a sample comprising leukocytes obtained from the subject, where the subject has been treated with an OX40 agonist, and where the one or more marker genes are selected from BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, H2-aa, H2-d, H2-k, ICOS, IL10, PDCA1, and TNFRSF18; and classifying the subject as responsive or non-responsive to the treatment based on the expression level of the one or more marker genes in the sample obtained from the subject, as compared with a reference, where an increased expression level of the one or more marker genes as compared with the reference indicates a responsive subject. These methods are based in part on the discovery described herein that expression of specific marker genes (e.g., BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, H2-aa, H2-d, H2-k, ICOS, IL10, PDCA1, and/or TNFRSF18) is upregulated following treatment with an OX40 agonist specifically in tumors that are responsive to such treatment. Expression level of a marker gene may be measured by one or more methods as described herein.

[0347] In some embodiments, a reference for monitoring responsiveness may include a sample from a subject not receiving a treatment (e.g., an OX40 agonist treatment). In some embodiments, a reference for monitoring responsiveness may include a sample from the same subject before receiving a treatment (e.g., an OX40 agonist treatment). In some embodiments, a reference for monitoring responsiveness may include a reference value from one or more samples of other patients receiving a treatment (e.g., an OX40 agonist treatment). For example, a population of patients may be treated, and a mean, average, or median value for expression level of one or more genes may be generated from the population as a whole. A set of samples obtained from cancers having a shared characteristic (e.g., the same cancer type and/or stage, or exposure to a common treatment such as an OX40 agonist) may be studied from a population, such as with a clinical outcome study. This set may be used to derive a reference, e.g., a reference number, to which a subject's sample may be compared. Any of the references described herein may be used as a reference for monitoring PD activity.

[0348] In some embodiments, the one or more marker genes are selected from CD80, CD86, ICOS, H2-aa, and CXCR3. In some embodiments, the marker gene may be one or more of the marker genes provided in Table 5 below. Table 5 provides a list of genes whose expression level was discovered as described herein to be upregulated in response OX40 agonist treatment in tumors that are responsive to such treatment (e.g., as a marker of

responsiveness). Exemplary gene/protein names or aliases, as well as an exemplary accession number corresponding to a human protein homolog, are provided.

Table 5.

NO.	Gene	Alias (non-exhaustive)	Accession No
1	BTLA	B- and T-lymphocyte attenuator; B- and T-lymphocyte-associated protein	NM_181780
2	CD4	T-cell surface glycoprotein CD4; CD4 receptor; CD4 antigen (p55); T-cell surface antigen T4/Leu-3	NM_000616
3	CD69	early activation antigen CD69; leukocyte surface antigen Leu-23; early T-cell activation antigen p60; early lymphocyte activation antigen; activation inducer molecule (AIM/CD69); C-type lectin domain family 2, member C	NM_001781
4	CD80	T-lymphocyte activation antigen CD80; activation B7-1 antigen; costimulatory factor CD80; CTLA-4 counter-receptor B7.1; B-lymphocyte activation antigen B7; costimulatory molecule variant IgV-CD80	NM_005191
5	CD83	CD83 antigen; B-cell activation protein; cell surface protein HB15; cell-surface glycoprotein	NM_004233
6	CD86	T-lymphocyte activation antigen CD86; BU63; FUN-1; CTLA-4 counter-receptor B7.2; B-lymphocyte activation antigen B7-2	NM_175862
7	CSF2	Granulocyte-macrophage colony-stimulating factor; GM-CSF; CSF;	NM_000749
8	CTLA4	Cytotoxic T-lymphocyte protein 4; CD152 isoform; celiac disease 3; insulin-dependent diabetes mellitus 12	NM_005214
9	CXCR3	C-X-C chemokine receptor type 3; Mig receptor; IP10 receptor; G protein-coupled receptor 9; interferon-inducible protein 10 receptor	NM_001504
10	Fcgr2b/CD32	low affinity immunoglobulin gamma Fc region receptor II-b; CDw32; Fc gamma RIIb	NM_004001
11	Fcgr3/CD16	low affinity immunoglobulin gamma Fc region receptor III-A; FcgammaRIIIA; CD16a antigen; neutrophil-specific antigen NA	NM_001127592
12	H2-aa	MHC Class IIA	NM_002122
13	H2-d	MHC Class I B	NM_005514
14	H2-k	MHC Class IA	NM_002116
15	ICOS	inducible T-cell costimulator; activation-inducible lymphocyte immunomediatory molecule	NM_012092
16	IL10	Interleukin-10; T-cell growth inhibitory factor; cytokine synthesis inhibitory factor	NM_000572
17	PDCA1	Bone marrow stromal cell antigen 2; NPC-A-7; HM1.24 antigen	NM_004335
18	TNFRSF18	Tumor necrosis factor receptor superfamily member 18; activation-inducible TNFR family receptor; glucocorticoid-induced TNFR-related protein; TNF receptor superfamily activation-inducible protein	NM_004195

[0349] In some embodiment, tumor may refer to a physical mass containing a plurality of cancer cells, *e.g.,* cells showing the characteristics of any of the cancers described herein. Examples of tumors may include primary tumors of any of the above types of cancer or metastatic tumors at a second site derived from any of the above types of cancer. In some embodiments, a tumor may contain cancer cells as well as tumor stroma.

[0350] Certain aspects of the present disclosure relate to measurement of the expression level of one or more genes in a sample. In some embodiments, a sample may include leukocytes. In some embodiments, the sample may be a tumor sample. A tumor sample may include cancer cells, lymphocytes, leukocytes, stroma, blood vessels, connective tissue, basal lamina, and any other cell type in association with the tumor. In some embodiments, the sample is a tumor tissue sample containing tumor-infiltrating leukocytes. For example, leukocyte associated with a tumor may be considered a tumor-infiltrating leukocyte. Examples of tumor-infiltrating leukocytes include without limitation T lymphocytes (such as CD8+ T lymphocytes and/or CD4+ T lymphocytes), B lymphocytes, or other bone marrow-lineage cells including granulocytes (neutrophils, eosinophils, basophils), monocytes, macrophages, dendritic cells (i.e., interdigitating dendritic cells), histiocytes, and natural killer cells. In some embodiments, a tumor-infiltrating leukocyte may be associated with cancer cells of a tumor. In some embodiments, a tumor-infiltrating leukocyte may be associated with tumor stroma.

[0351] In some embodiments, the sample may be processed to separate or isolate one or more cell types (e.g., leukocytes). In some embodiments, the sample may be used without separating or isolating cell types. A tumor sample may be obtained from a subject by any method known in the art, including without limitation a biopsy, endoscopy, or surgical procedure. In some embodiments, a tumor sample may be prepared by methods such as freezing, fixation (e.g., by using formalin or a similar fixative), and/or embedding in paraffin wax. In some embodiments, a tumor sample may be sectioned. In some embodiments, a fresh tumor sample (i.e., one that has not been prepared by the methods described above) may be used. In some embodiments, a sample may be prepared by incubation in a solution to preserve mRNA and/or protein integrity. A tumor sample containing leukocytes may be assayed by any technique described herein for measuring marker gene expression level. In some embodiments, the tumor samples are enriched for tumor area by macrodissection.

[0352] In some embodiments, the sample may be a peripheral blood sample. A peripheral blood sample may include white blood cells, PBMCs, and the like. Any technique known in the art for isolating leukocytes from a peripheral blood sample may be used. For example, a blood sample may be drawn, red blood cells may be lysed, and a white blood cell pellet may be isolated and used for the sample. In another example, density gradient separation may be used to separate leukocytes (e.g., PBMCs) from red blood cells. In some embodiments, a fresh peripheral blood sample (i.e., one that has not been prepared by the methods described above) may be used. In some embodiments, a peripheral blood sample may be prepared by incubation in a solution to preserve mRNA and/or protein integrity.

[0353] In some embodiments, responsiveness to treatment may refer to any one or more of: extending survival (including overall survival and progression free survival); resulting in an objective response (including a complete response or a partial response); or improving signs or symptoms of cancer. In some embodiments, responsiveness may refer to improvement of one or more factors according to the published set of RECIST guidelines for determining the status of a tumor in a cancer patient, *i.e.,* responding, stabilizing, or progressing. For a more detailed discussion of these guidelines, see Eisenhauer et al., *Eur J Cancer* 2009;45: 228–47; Topalian et al., *N Engl J Med* 2012;366:2443–54; Wolchok et al., *Clin Can Res* 2009;15:7412–20; and Therasse, P., *et al. J. Natl. Cancer Inst.* 92:205-16 (2000). A responsive subject may refer to a subject whose cancer(s) show improvement, e.g., according to one or more factors based on RECIST criteria. A non-responsive subject may refer to a subject whose cancer(s) do not show improvement, e.g., according to one or more factors based on RECIST criteria.

[0354] Conventional response criteria may not be adequate to characterize the anti-tumor activity of immunotherapeutic agents, which can produce delayed responses that may be preceded by initial apparent radiological progression, including the appearance of new lesions. Therefore, modified response criteria have been developed that account for the possible appearance of new lesions and allow radiological progression to be confirmed at a subsequent assessment. Accordingly, in some embodiments, responsiveness may refer to improvement of one of more factors according to immune-related response criteria2(irRC). See, e.g., Wolchok et al., Clin Can Res 2009;15:7412 – 20. In some embodiments, new lesions are added into the defined tumor burden and followed, e.g., for radiological progression at a subsequent assessment. In some embodiments, presence of non-target lesions are included in assessment of complete response and not included in assessment of radiological progression. In some embodiments,

radiological progression may be determined only on the basis of measurable disease and/or may be confirmed by a consecutive assessment ≥ 4 weeks from the date first documented.

[0355] In some embodiments, responsiveness may include immune activation. In some embodiments, responsiveness may include treatment efficacy. In some embodiments, responsiveness may include immune activation and treatment efficacy.

VI. Methods of Treatment

[0356] In one aspect, provided herein are methods for treating or delaying progression of cancer in an individual comprising administering to the individual an effective amount of an OX40 agonist. The methods of this disclosure may find use, *inter alia*, in treating conditions where enhanced immunogenicity is desired such as increasing tumor immunogenicity for the treatment of cancer or T cell dysfunctional disorders. A variety of cancers may be treated, or their progression may be delayed, by these methods.

[0357] Provided herein are methods for treating or delaying progression of cancer in a subject by measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the one or more marker genes are selected from CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and if the expression level of said one or more marker genes in the sample obtained from the subject is higher than a reference, administering to the subject an effective amount of an OX40 agonist. Further provided herein are methods for treating or delaying progression of cancer in a subject including administering to the subject an effective amount of an OX40 agonist, where a sample containing leukocytes obtained from the subject has increased expression of one or more marker genes are selected from CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R, as compared with a reference. These methods are based in part on the discovery described herein that higher expression of certain immune activation and Th1 markers (e.g., CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and/or IL7R) is associated with better responsiveness to OX40 agonist treatment.

[0358] Yet further provided herein are methods for treating or delaying progression of cancer in a subject by measuring the expression level of one or more marker genes in a sample containing leukocytes obtained from the subject, where the one or more marker genes

are selected from CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and if the expression level of said one or more marker genes in the sample obtained from the subject is lower than a reference, administering to the subject an effective amount of an OX40 agonist. Further provided herein are methods for treating or delaying progression of cancer in a subject including administering to the subject an effective amount of an OX40 agonist, where a sample containing leukocytes obtained from the subject has decreased expression of one or more marker genes are selected from CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1, as compared with a reference. These methods are based in part on the discovery described herein that lower expression of certain immune activation and Th1 markers (e.g., CSF2, CCL22, EPCAM, GATA3, IL13, and/or VTCN1) is associated with better responsiveness to OX40 agonist treatment.

[0359] In some embodiments, the OX40 agonist is administered to a subject wherein a sample containing leukocytes from the subject have been detected for increased expression of one or more marker genes selected from the group consisting of CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R, and/or decreased expression of one or more marker selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1.

[0360] A sample or cell that expresses a protein of interest (such as CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, IL7R, CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1) may be one in which mRNA encoding the protein, or the protein, including fragments thereof, is determined to be present in the sample or cell.

[0361] A sample, cell, tumor, or cancer which has increased expression of one or more markers (such as CD8a, CD8b, H2-d, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, IL7R) in a type of cancer may be one in which the level of one or more marker expression may be considered increased to a skilled person for that type of cancer. For example, such increase may be at least about 0.5 fold, at least about 1 fold, at least about 2 fold, or at least about 5 fold relative to the levels in a population of samples, cells, tumors, or cancers of the same cancer type.

[0362] A sample, cell, tumor, or cancer which has decreased expression of one or more markers (such as CSF2, CCL22, EPCAM, GATA3, IL13, and/or VTCN1) in a type of cancer may be one in which the level of one or more marker expression may be considered decreased to a skilled person for that type of cancer. For example, such decrease may be at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about

60%, at least about 70% decrease relative to the levels in a population of samples, cells, tumors, or cancers of the same cancer type.

[0363] In some embodiments, a cancer to be treated by the methods of the present disclosure includes, but is not limited to, squamous cell cancer (*e.g.*, epithelial squamous cell cancer), lung cancer including small-cell lung cancer, non-small cell lung cancer, adenocarcinoma of the lung and squamous carcinoma of the lung, cancer of the peritoneum, hepatocellular cancer, gastric or stomach cancer including gastrointestinal cancer and gastrointestinal stromal cancer, pancreatic cancer, glioblastoma, cervical cancer, ovarian cancer, liver cancer, bladder cancer, cancer of the urinary tract, hepatoma, breast cancer, colon cancer, rectal cancer, colorectal cancer, endometrial or uterine carcinoma, salivary gland carcinoma, kidney or renal cancer, prostate cancer, vulval cancer, thyroid cancer, hepatic carcinoma, anal carcinoma, penile carcinoma, melanoma, superficial spreading melanoma, lentigo maligna melanoma, acral lentiginous melanomas, nodular melanomas, multiple myeloma and B-cell lymphoma; chronic lymphocytic leukemia (CLL); acute lymphoblastic leukemia (ALL); hairy cell leukemia; chronic myeloblastic leukemia; and post-transplant lymphoproliferative disorder (PTLD), as well as abnormal vascular proliferation associated with phakomatoses, edema (such as that associated with brain tumors), Meigs' syndrome, brain, as well as head and neck cancer, and associated metastases. In some embodiments, the cancer is colorectal cancer. In some embodiments, the cancer is selected from non-small cell lung cancer, renal cell carcinoma, ovarian cancer, bladder cancer, glioblastoma, neuroblastoma, melanoma, breast carcinoma, gastric cancer, and hepatocellular carcinoma. In some embodiments, the cancer is triple-negative breast carcinoma. In some embodiments, the cancer may be an early stage cancer or a late stage cancer. In some embodiments, the cancer may be a primary tumor. In some embodiments, the cancer may be a metastatic tumor at a second site derived from any of the above types of cancer.

[0364] In some embodiments, examples of cancer further include, but are not limited to, B-cell lymphoma (including low grade/follicular non-Hodgkin's lymphoma (NHL); small lymphocytic (SL) NHL; intermediate grade/follicular NHL; intermediate grade diffuse NHL; high grade immunoblastic NHL; high grade lymphoblastic NHL; high grade small non-cleaved cell NHL; bulky disease NHL; mantle cell lymphoma; AIDS-related lymphoma; and Waldenstrom's Macroglobulinemia); chronic lymphocytic leukemia (CLL); acute lymphoblastic leukemia (ALL); Hairy cell leukemia; chronic myeloblastic leukemia; and post-transplant lymphoproliferative disorder (PTLD), as well as abnormal vascular

proliferation associated with phakomatoses, edema (such as that associated with brain tumors), B-cell proliferative disorders, and Meigs' syndrome. More specific examples include, but are not limited to, relapsed or refractory NHL, front line low grade NHL, Stage III/IV NHL, chemotherapy resistant NHL, precursor B lymphoblastic leukemia and/or lymphoma, small lymphocytic lymphoma, B-cell chronic lymphocytic leukemia and/or prolymphocytic leukemia and/or small lymphocytic lymphoma, B-cell prolymphocytic lymphoma, immunocytoma and/or lymphoplasmacytic lymphoma, lymphoplasmacytic lymphoma, marginal zone B-cell lymphoma, splenic marginal zone lymphoma, extranodal marginal zone—MALT lymphoma, nodal marginal zone lymphoma, hairy cell leukemia, plasmacytoma and/or plasma cell myeloma, low grade/follicular lymphoma, intermediate grade/follicular NHL, mantle cell lymphoma, follicle center lymphoma (follicular), intermediate grade diffuse NHL, diffuse large B-cell lymphoma, aggressive NHL (including aggressive front-line NHL and aggressive relapsed NHL), NHL relapsing after or refractory to autologous stem cell transplantation, primary mediastinal large B-cell lymphoma, primary effusion lymphoma, high grade immunoblastic NHL, high grade lymphoblastic NHL, high grade small non-cleaved cell NHL, bulky disease NHL, Burkitt's lymphoma, precursor (peripheral) large granular lymphocytic leukemia, mycosis fungoides and/or Sezary syndrome, skin (cutaneous) lymphomas, anaplastic large cell lymphoma, angiocentric lymphoma.

[0365] In some embodiments, examples of cancer further include, but are not limited to, B-cell proliferative disorders, which further include, but are not limited to, lymphomas (e.g., B-Cell Non-Hodgkin's lymphomas (NHL)) and lymphocytic leukemias. Such lymphomas and lymphocytic leukemias include e.g. a) follicular lymphomas, b) Small Non-Cleaved Cell Lymphomas/ Burkitt's lymphoma (including endemic Burkitt's lymphoma, sporadic Burkitt's lymphoma and Non-Burkitt's lymphoma), c) marginal zone lymphomas (including extranodal marginal zone B-cell lymphoma (Mucosa-associated lymphatic tissue lymphomas, MALT), nodal marginal zone B-cell lymphoma and splenic marginal zone lymphoma), d) Mantle cell lymphoma (MCL), e) Large Cell Lymphoma (including B-cell diffuse large cell lymphoma (DLCL), Diffuse Mixed Cell Lymphoma, Immunoblastic Lymphoma, Primary Mediastinal B-Cell Lymphoma, Angiocentric Lymphoma-Pulmonary B-Cell Lymphoma), f) hairy cell leukemia, g) lymphocytic lymphoma, Waldenstrom's macroglobulinemia, h) acute lymphocytic leukemia (ALL), chronic lymphocytic leukemia (CLL)/ small lymphocytic lymphoma (SLL), B cell prolymphocytic leukemia, i) plasma cell neoplasms, plasma cell myeloma, multiple myeloma, plasmacytoma, and/or j) Hodgkin's disease.

[0366] In some embodiments of any of the methods, the cancer is a B-cell proliferative disorder. In some embodiments, the B-cell proliferative disorder is lymphoma, non-Hodgkins lymphoma (NHL), aggressive NHL, relapsed aggressive NHL, relapsed indolent NHL, refractory NHL, refractory indolent NHL, chronic lymphocytic leukemia (CLL), small lymphocytic lymphoma, leukemia, hairy cell leukemia (HCL), acute lymphocytic leukemia (ALL), or mantle cell lymphoma. In some embodiments, the B-cell proliferative disorder is NHL, such as indolent NHL and/or aggressive NHL. In some embodiments, the B-cell proliferative disorder is indolent follicular lymphoma or diffuse large B-cell lymphoma.

[0367] In some embodiments, the subject has cancer or is at risk of developing cancer. In some embodiments, the treatment results in a sustained response in the subject after cessation of the treatment. In some embodiments, the subject has cancer that may be at early stage or late stage. In some embodiments, the subject is a human. In some embodiments, the subject is a mammal, such as domesticated animals (e.g., cows, sheep, cats, dogs, and horses), primates (e.g., humans and non-human primates such as monkeys), rabbits, and rodents (e.g., mice and rats).

[0368] In some embodiments, provided is a method for treating or delaying progression of cancer in a subject comprising administering to the subject an effective amount of an OX40 agonist, and further comprising administering an additional therapy. The additional therapy may be radiation therapy, surgery (*e.g.*, lumpectomy and a mastectomy), chemotherapy, gene therapy, DNA therapy, viral therapy, RNA therapy, immunotherapy, bone marrow transplantation, nanotherapy, monoclonal antibody therapy, or a combination of the foregoing. The additional therapy may be in the form of adjuvant or neoadjuvant therapy. In some embodiments, the additional therapy is the administration of small molecule enzymatic inhibitor or anti-metastatic agent. In some embodiments, the additional therapy is the administration of side-effect limiting agents (*e.g.,* agents intended to lessen the occurrence and/or severity of side effects of treatment, such as anti-nausea agents, etc.). In some embodiments, the additional therapy is radiation therapy. In some embodiments, the additional therapy is surgery. In some embodiments, the additional therapy is a combination of radiation therapy and surgery. In some embodiments, the additional therapy is gamma irradiation. In some embodiments, the additional therapy is therapy targeting PI3K/AKT/mTOR pathway, HSP90 inhibitor, tubulin inhibitor, apoptosis inhibitor, and/or chemopreventative agent. The additional therapy may be one or more of the chemotherapeutic agents described hereabove.

[0369] Such combination therapies noted above encompass combined administration (where two or more therapeutic agents are included in the same or separate formulations), and separate administration, in which case, administration of the OX40 agonist of the invention can occur prior to, simultaneously, and/or following, administration of the additional therapeutic agent or agents. In one embodiment, administration of the OX40 agonist and administration of an additional therapeutic agent occur within about one month, or within about one, two or three weeks, or within about one, two, three, four, five, or six days, of each other. OX40 agonists of the invention can also be used in combination with radiation therapy.

[0370] In some embodiments, an anti-human OX40 agonist may be administered in conjunction with a chemotherapy or chemotherapeutic agent. In some embodiments, an anti-human OX40 agonist may be administered in conjunction with a radiation therapy or radiotherapeutic agent. In some embodiments, an anti-human OX40 agonist may be administered in conjunction with a targeted therapy or targeted therapeutic agent. In some embodiments, an anti-human OX40 agonist may be administered in conjunction with an immunotherapy or immunotherapeutic agent, for example a monoclonal antibody.

[0371] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with in combination with a PD-1 axis binding antagonist. A PD-1 axis binding antagonist includes but is not limited to a PD-1 binding antagonist, a PD-L1 binding antagonist and a PD-L2 binding antagonist. Alternative names for "PD-1" include CD279 and SLEB2. Alternative names for "PD-L1" include B7-H 1, B7-4, CD274, and B7-H. Alternative names for "PD-L2" include B7-DC, Btdc, and CD273. In some embodiments, PD-1, PD-Ll , and PD-L2 are human PD-1 , PD-Ll and PD-L2. In some embodiments, the PD-1 binding antagonist is a molecule that inhibits the binding of PD-1 to its ligand binding partners. In a specific aspect the PD-1 ligand binding partners are PD-Ll and/or PD-L2. In another embodiment, a PD-Ll binding antagonist is a molecule that inhibits the binding of PD-Ll to its binding partners. In a specific aspect, PD-Ll binding partners are PD-1 and/or B7- 1. In another embodiment, the PD-L2 binding antagonist is a molecule that inhibits the binding of PD-L2 to its binding partners. In a specific aspect, a PD-L2 binding partner is PD-1. The antagonist may be an antibody, an antigen binding fragment thereof, an immunoadhesin, a fusion protein, or oligopeptide. In some embodiment, the PD-1 binding antagonist is an anti-PD-1 antibody (*e.g.*, a human antibody, a humanized antibody, or a chimeric antibody). In some embodiments, the anti-PD-1 antibody is selected from the group consisting of MDX-1 106, Merck 3475 and CT- 011. In some embodiments, the PD-1

binding antagonist is an immunoadhesin (*e.g.*, an immunoadhesin comprising an extracellular or PD-1 binding portion of PD-L1 or PD-L2 fused to a constant region (*e.g.*, an Fc region of an immunoglobulin sequence). In some embodiments, the PD-1 binding antagonist is AMP-224. In some embodiments, the PD-Ll binding antagonist is an anti-PD-Ll antibody. In some embodiments, the anti-PD-Ll binding antagonist is selected from the group consisting of YW243.55.S70, MPDL3280A (atezolizumab), MEDI4736 (durvalumab), MDX-1105, and MSB0010718C (avelumab). MDX- 1 105, also known as BMS-936559, is an anti-PD-Ll antibody described in WO2007/005874. Antibody YW243.55.S70 (heavy and light chain variable region sequences shown in SEQ ID Nos. 20 and 21, respectively) is an anti-PD-Ll described in WO 2010/077634 Al . MDX-1 106, also known as MDX-1 106-04, ONO-4538 or BMS-936558, is an anti-PD-1 antibody described in WO2006/121168. Merck 3745, also known as MK-3475, SCH-900475, pembrolizumab, and KEYTRUDA®, is an anti-PD-1 antibody described in WO2009/114335. CT-011, also known as hBAT, hBAT-1, and pidilizumab, is an anti- PD-1 antibody described in WO2009/101611. AMP-224, also known as B7-DCIg, is a PD-L2- Fc fusion soluble receptor described in WO2010/027827 and WO201 1/066342. In some embodiments, the anti-PD-1 antibody is MDX- 1 106. Alternative names for "MDX- 1106" include MDX-1 106-04, ONO-4538, BMS-936558, Nivolumab, or OPDIVO®. In some embodiments, the anti-PD-1 antibody is Nivolumab (CAS Registry Number: 946414-94-4). In some embodiments, the anti-PD-1 antibody is selected from the group consisting of MDX-1106 (nivolumab, OPDIVO®), Merck 3475 (MK-3475, pembrolizumab, KEYTRUDA®), CT- 011 (Pidilizumab), MEDI-0680 (AMP-514), PDR001, REGN2810, BGB-108, and BGB-A317.

[0372] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antagonist directed against CTLA-4 (also known as CD152), *e.g.,* a blocking antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with ipilimumab (also known as MDX-010, MDX-101, or Yervoy®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with tremelimumab (also known as ticilimumab or CP-675,206). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antagonist directed against B7-H3 (also known as CD276), *e.g.,* a blocking antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with MGA271. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be

administered in conjunction with an antagonist directed against a TGF beta, *e.g.,* metelimumab (also known as CAT-192), fresolimumab (also known as GC1008), or LY2157299.

[0373] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment comprising adoptive transfer of a T cell (*e.g.,* a cytotoxic T cell or CTL) expressing a chimeric antigen receptor (CAR). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment comprising adoptive transfer of a T cell comprising a dominant-negative TGF beta receptor, *e.g,* a dominant-negative TGF beta type II receptor. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment comprising a HERCREEM protocol (see, e.g., ClinicalTrials.gov Identifier NCT00889954).

[0374] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an agonist directed against CD137 (also known as TNFRSF9, 4-1BB, or ILA), *e.g.,* an activating antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with urelumab (also known as BMS-663513). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an agonist directed against CD40, *e.g.,* an activating antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with CP-870893. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an agonist directed against OX40 (also known as CD134), *e.g.,* an activating antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a different anti-OX40 antibody (e.g., AgonOX). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an agonist directed against CD27, *e.g.,* an activating antibody. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with CDX-1127. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antagonist directed against indoleamine-2,3-dioxygenase (IDO). In some embodiments, with the IDO antagonist is 1-methyl-D-tryptophan (also known as 1-D-MT).

[0375] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody-drug conjugate. In some

embodiments, the antibody-drug conjugate comprises mertansine or monomethyl auristatin E (MMAE). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with and anti-NaPi2b antibody-MMAE conjugate (also known as DNIB0600A or RG7599). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with trastuzumab emtansine (also known as T-DM1, ado-trastuzumab emtansine, or KADCYLA®, Genentech). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with DMUC5754A. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody-drug conjugate targeting the endothelin B receptor (EDNBR), *e.g.,* an antibody directed against EDNBR conjugated with MMAE.

[0376] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an angiogenesis inhibitor. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody directed against a VEGF, *e.g.,* VEGF-A. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with bevacizumab (also known as AVASTIN®, Genentech). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody directed against angiopoietin 2 (also known as Ang2). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with MEDI3617.

[0377] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antineoplastic agent. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an agent targeting CSF-1R (also known as M-CSFR or CD115). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with anti-CSF-1R (also known as IMC-CS4). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an interferon, for example interferon alpha or interferon gamma. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with Roferon-A (also known as recombinant Interferon alpha-2a). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with GM-CSF (also known as recombinant human granulocyte macrophage colony stimulating factor, rhu GM-CSF,

sargramostim, or Leukine®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with IL-2 (also known as aldesleukin or Proleukin®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with IL-12. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody targeting CD20. In some embodiments, the antibody targeting CD20 is obinutuzumab (also known as GA101 or Gazyva®) or rituximab. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an antibody targeting GITR. In some embodiments, the antibody targeting GITR is TRX518.

[0378] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a cancer vaccine. In some embodiments, the cancer vaccine is a peptide cancer vaccine, which in some embodiments is a personalized peptide vaccine. In some embodiments the peptide cancer vaccine is a multivalent long peptide, a multi-peptide, a peptide cocktail, a hybrid peptide, or a peptide-pulsed dendritic cell vaccine (see, e.g., Yamada et al., Cancer Sci, 104:14-21, 2013). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an adjuvant. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment comprising a TLR agonist, *e.g.,* Poly-ICLC (also known as Hiltonol®), LPS, MPL, or CpG ODN. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with tumor necrosis factor (TNF) alpha. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with IL-1. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with HMGB1. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an IL-10 antagonist. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an IL-4 antagonist. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an IL-13 antagonist. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an HVEM antagonist. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an ICOS agonist, *e.g.,* by administration of ICOS-L, or an agonistic antibody directed against ICOS. In some

embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment targeting CX3CL1. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment targeting CXCL9. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment targeting CXCL10. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a treatment targeting CCL5. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an LFA-1 or ICAM1 agonist. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a Selectin agonist.

[0379] In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a targeted therapy. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of B-Raf. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with vemurafenib (also known as Zelboraf®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with dabrafenib (also known as Tafinlar®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with erlotinib (also known as Tarceva®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of a MEK, such as MEK1 (also known as MAP2K1) or MEK2 (also known as MAP2K2). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with cobimetinib (also known as GDC-0973 or XL-518). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with trametinib (also known as Mekinist®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of K-Ras. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of c-Met. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with onartuzumab (also known as MetMAb). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of Alk. In some embodiments, an OX40

agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with AF802 (also known as CH5424802 or alectinib). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of a phosphatidylinositol 3-kinase (PI3K). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with BKM120. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with idelalisib (also known as GS-1101 or CAL-101). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with perifosine (also known as KRX-0401). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of an Akt. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with MK2206. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with GSK690693. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with GDC-0941. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with an inhibitor of mTOR. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with sirolimus (also known as rapamycin). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with temsirolimus (also known as CCI-779 or Torisel®). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with everolimus (also known as RAD001). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with ridaforolimus (also known as AP-23573, MK-8669, or deforolimus). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with OSI-027. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with AZD8055. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with INK128. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with a dual PI3K/mTOR inhibitor. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with XL765. In some embodiments, an OX40 agonist (e.g., an anti-human

OX40 agonist antibody) may be administered in conjunction with GDC-0980. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with BEZ235 (also known as NVP-BEZ235). In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with BGT226. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with GSK2126458. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with PF-04691502. In some embodiments, an OX40 agonist (e.g., an anti-human OX40 agonist antibody) may be administered in conjunction with PF-05212384 (also known as PKI-587).

[0380] An OX40 agonist of the invention (and any additional therapeutic agent) can be administered by any suitable means, including parenteral, intrapulmonary, and intranasal, and, if desired for local treatment, intralesional administration. Parenteral infusions include intramuscular, intravenous, intraarterial, intraperitoneal, or subcutaneous administration. Dosing can be by any suitable route, e.g. by injections, such as intravenous or subcutaneous injections, depending in part on whether the administration is brief or chronic. Various dosing schedules including but not limited to single or multiple administrations over various time-points, bolus administration, and pulse infusion are contemplated herein.

[0381] OX40 agonists of the invention would be formulated, dosed, and administered in a fashion consistent with good medical practice. Factors for consideration in this context include the particular disorder being treated, the particular mammal being treated, the clinical condition of the individual patient, the cause of the disorder, the site of delivery of the agent, the method of administration, the scheduling of administration, and other factors known to medical practitioners. The antibody need not be, but is optionally formulated with one or more agents currently used to prevent or treat the disorder in question. The effective amount of such other agents depends on the amount of antibody present in the formulation, the type of disorder or treatment, and other factors discussed above. These are generally used in the same dosages and with administration routes as described herein, or about from 1 to 99% of the dosages described herein, or in any dosage and by any route that is empirically/clinically determined to be appropriate.

[0382] For the prevention or treatment of disease, the appropriate dosage of an OX40 agonist of the invention (when used alone or in combination with one or more other additional therapeutic agents) will depend on the type of disease to be treated, the type of antibody, the severity and course of the disease, whether the antibody is administered for

preventive or therapeutic purposes, previous therapy, the patient's clinical history and response to the antibody, and the discretion of the attending physician. The antibody is suitably administered to the patient at one time or over a series of treatments. Depending on the type and severity of the disease, about 1 μg/kg to 40 mg/kg of antibody can be an initial candidate dosage for administration to the patient, whether, for example, by one or more separate administrations, or by continuous infusion. One typical daily dosage might range from about 1 μg/kg to 100 mg/kg or more, depending on the factors mentioned above. For repeated administrations over several days or longer, depending on the condition, the treatment would generally be sustained until a desired suppression of disease symptoms occurs. Such doses may be administered intermittently, e.g. every week or every three weeks (*e.g.* such that the patient receives from about two to about twenty, or *e.g.* about six doses of the antibody). An initial higher loading dose, followed by one or more lower doses may be administered. However, other dosage regimens may be useful. The progress of this therapy is easily monitored by conventional techniques and assays.

[0383] In some embodiments of the methods of the present disclosure, the cancer has elevated levels of T cell infiltration. As used herein, T cell infiltration of a cancer may refer to the presence of T cells, such as tumor-infiltrating lymphocytes (TILs), within or otherwise associated with the cancer tissue. It is known in the art that T cell infiltration may be associated with improved clinical outcome in certain cancers (see, *e.g.*, Zhang *et al.*, N. Engl. J. Med. 348(3):203-213 (2003)). In some embodiments, the TILs may be OX40+. In some embodiments, the TILs may be CD4+ OX40+ Foxp3+ Treg or CD4+ OX40+ Foxp3- Teff cells.

VII. Kits and Articles of Manufacture

[0384] For use in the applications described or suggested above, kits or articles of manufacture are also provided by the invention. Such kits may comprise at least one reagent specific for detecting expression level of a marker gene described herein (e.g., genes described in Tables 3-5), and may further include instructions for carrying out a method described herein.

[0385] In some embodiments, the invention provides compositions and kits comprising primers and primer pairs, which allow the specific amplification of the polynucleotides of the marker genes or of any specific parts thereof, and probes that selectively or specifically hybridize to nucleic acid molecules described herein or to any part thereof. Probes may be

labeled with a detectable marker, such as, for example, a radioisotope, fluorescent compound, bioluminescent compound, a chemiluminescent compound, metal chelator or enzyme. Such probes and primers can be used to detect the presence of polynucleotides, such as the polynucleotides corresponding to genes listed in Tables 3-5, in a sample and as a means for detecting cell expressing proteins encoded by the polynucleotides. As will be understood by the skilled artisan, a great many different primers and probes may be prepared based on the sequences provided herein and used effectively to amplify, clone and/or determine the presence and/or levels of mRNAs. In some embodiments, the kits may further comprise a surface or substrate (such as a microarray) for capture probes for detecting of amplified nucleic acids. In some embodiments, the kits comprise at least one pair of primers and a probe specific for detecting one marker gene expression level using qRT-PCR.

[0386] The reagents for detecting the protein expression level of a marker gene may comprise an antibody that specifically binds to the protein encoded by the marker gene.

[0387] In some embodiments, the kits further comprise an OX40 agonist (e.g., an anti-OX40 agonist antibody). The kits may further comprise instructions to administering an OX40 agonist if the patient is identified as responsive to the OX40 agonist treatment.

[0388] The kits may further comprise a carrier means being compartmentalized to receive in close confinement one or more container means such as vials, tubes, and the like, each of the container means comprising one of the separate elements to be used in the method. For example, one of the container means may comprise a probe that is or can be detectably labeled. Such probe may be an antibody or polynucleotide specific for a marker gene. Where the kit utilizes nucleic acid hybridization to detect the target nucleic acid, the kit may also have containers containing nucleotide(s) for amplification of the target nucleic acid sequence and/or a container comprising a reporter-means, such as a biotin-binding protein, such as avidin or streptavidin, bound to a reporter molecule, such as an enzymatic, florescent, or radioisotope label.

[0389] The kit of the invention will typically comprise the container described above and one or more other containers comprising materials desirable from a commercial and user standpoint, including buffers, diluents, filters, needles, syringes, and package inserts with instructions for use. A label may be present on the container to indicate that the composition is used for a specific therapy or non-therapeutic application, and may also indicate directions for either *in vivo* or *in vitro* use, such as those described above.

[0390] The kit can further comprise a set of instructions and materials for preparing a tissue or cell sample and preparing nucleic acids (such as mRNA) from the sample.

[0391] The specification is considered to be sufficient to enable one skilled in the art to practice the invention. Various modifications of the invention in addition to those shown and described herein will become apparent to those skilled in the art from the foregoing description and fall within the scope of the appended claims.

[0392] All publications, patents, and patent applications cited herein are hereby incorporated by reference in their entirety for all purposes.

EXAMPLES

[0393] The invention can be further understood by reference to the following examples, which are provided by way of illustration and are not meant to be limiting.

Example 1: OX40 agonist treatment modulates T cell activities and anti-tumor activity

[0394] OX40 is known to be a co-stimulatory molecule expressed on activated CD4 T cells (Teff) and T regulatory (Treg) cells. Ligation of OX40 in the presence of TCR stimulation is known to enhance T effector cell function via dual mechanism of potentiating activation of Teff cells and inhibiting Treg cells.

[0395] Enhancement of anti-tumor immunity by OX40 agonist treatment is a promising therapeutic approach to treat cancer. Therefore, *in vitro* and *in vivo* preclinical studies were conducted to characterize co-stimulatory activity, evaluate anti-tumor efficacy, and identify predictive and pharmacodynamic (PD) biomarkers of OX40 agonists.

Materials and Methods

In vitro *T cell co-stimulation assay*

[0396] $CD4^+$ T cells were isolated from PBMCs from healthy donors via magnetic enrichment (Miltenyi). Sorted $CD4^+$ were stimulated with PHA to induce OX40 expression, followed by resting in the presence of IL-2, and subsequently restimulated with a fixed concentration of anti-CD3 and variable anti-OX40 (anti-human OX40 mAb) concentrations in platebound form. T cell proliferation was measured by quantitation of ATP levels (Promega), and supernatant IFN-gamma levels were assessed by ELISA.

In vitro *Treg suppression assay*

[0397] $CD4^+CD25^+CD127^-$ Tregs and naïve T cells were isolated from healthy donors via FACS and magnetic purification, respectively. Suppression assay cultures were comprised of Tregs and CFSE labeled naïve $CD4^+$ cells (2 Treg:1 naive), irradiated $CD80^+$ $CD32^+$ L cells

as APCs, anti-CD3, and anti-human OX40 mAb. Suppression of naïve T cell proliferation was quantified via FACS.

In vivo tumor studies

[0398] Syngeneic tumor cells were inoculated either subcutaneously (CT26, 51Blim10) or orthotopically (EMT6, JC) in C57/Bl6 or BalbC mice. For predictive biomarker analysis, tumors were harvested when they reached a volume of 150-250 mm^3. For efficacy and pharmacodynamic biomarker analysis, when the tumors reached a volume of 150-250 mm^3, mice were treated with 10 mg/kg of either control Ab or murine anti-mouse OX40 mAb 2x/wk for 3 weeks. Tumors and peripheral blood were harvested on day 2, day 9, and day 16 post first dose.

Flow cytometry

[0399] Tumor tissues were cut into small pieces, incubated with collagenase and DNAse (Roche), and single cell suspensions were prepared using a gentle MACS Dissociator (Miltenyi) per manufacturer's protocol. Phenotyping of tumor infiltrating lymphocytes (TILs) was performed with commercial antibodies against CD45, CD3, CD4, CD8, CD25, (all BD Biosciences) and Foxp3 (eBiosciences) per manufacturers' instructions. The Live/Dead Fixable Near-IR Dead Cell Stain Kit (Life Technologies) protocol was used to exclude dead cells from analysis. Stained cells were analyzed on a BD FACSCanto flow cytometer.

Gene expression analysis

[0400] RNA was extracted from tumor lysates using the mirvana miRNA Isolation kit (Life Technologies) according to the total RNA isolation protocol supplied by the manufacturer. RNA quality was assessed using an Agilent 2100 Bioanalyzer. RNA (200 ng) was subjected to a one-step cDNA synthesis/preamplification reaction using the Invitrogen Platinum Taq/Reverse Transcriptase enzyme mix as per the manufacturer's protocol with the exception that PCR cycling conditions were changed from a 14 cycle preamplification to 16 cycles (Life Technologies). Following amplification, samples were diluted one to four with TE and qPCR was conducted on Fluidigm 96.96 Dynamic Arrays using the BioMark™ HD system according to the manufacturer's protocol and as previously described (Shames, D.S., *et al.* (2013) *PLoS ONE* 8:e56765). Samples were run in duplicate and cycle threshold (Ct) values were converted to relative expression values (negative delta Ct) by subtracting the mean of

the three reference genes from the mean of each target gene. A list of all genes evaluated in these studies is provided in Table 6 below.

Table 6

Assay	Gene	Assay	Gene	Assay	Gene	Assay	Gene
1	CD3E	25	CD274	49	CCL5	73	H2-k
2	CD4	26	CD276	50	CCL22	74	Fcgr2b/CD32
3	CD8A	27	PDCD1LG2	51	PVR	75	ICAM1
4	CD8b	28	VTCN1	52	CCR5	76	VCAM1
5	GATA3	29	IL1B	53	CCR7	77	EPCAM
6	TBX21	30	IL2	54	CX3CL1	78	TNFSF4 (OX40L)
7	RORC	31	IL4	55	CXCR3	79	TNFSF9 (4-1BBL)
8	FOXP3	32	IL6	56	VEGFA	80	TNFRSF9 (4-1BB)
9	BTLA	33	IL7	57	CD40	81	HAVCR2
10	CTLA4	34	IL7R	58	MS4A1	82	TNFRSF14
11	PDCD1	35	IL10	59	Fcgr3/CD16	83	FasL
12	CD28	36	IL12A	60	KLRK1	84	CD48
13	CD27	37	IL13	61	TIGIT	85	TNFRSF18 (GITR)
14	CD69	38	IL17A	62	IFNa	86	PTPRC
15	ICOS	39	IL33	63	NCAM1	87	PGE2/PGES2
16	EOMES	40	CSF2	64	CD83	88	PTGER4
17	CD226	41	IFNg	65	CD209a	89	IDO1
18	LAG3	42	TGFB1	66	PDCA1	90	LGALS9
19	TNFRSF4	43	TNF	67	Zbtb46	91	MAP4K1
20	GZMA	44	IL2RA	68	PTGS2	92	IL9
21	GZMB	45	CXCL1	69	ARG1	93	SP2
22	PRF1	46	CXCL9	70	ITGAM	94	YWHAZ
23	CD80	47	CXCL10	71	H2-d	95	GUSb
24	CD86	48	CCL2	72	H2-aa	96	Fcgr1/CD64

Data analysis

[0401] For predictive biomarker evaluation, expression of each gene was normalized to median expression. Genes that showed at least an average of 2 fold statistically significant (Student's t-test, $p<0.05$) differential expression between more responsive (EMT6 and CT26) and less responsive (JC and 51Blim10) models were selected for downstream analysis. Hierarchical clustering was carried out on median-centered data with the complete linkage

method using Cluster v3.0 and visualized using Treeview (Eisen, M.B., *et al.* (1998) *Proc. Natl. Acad. Sci.* 95:14863-8).

Results

[0402] OX40 is known to be highly expressed on Teff and Treg cells in mouse and human tumors. As such, modulating OX40 activity may provide a means to modulate T cell function in cancer, *i.e.,* for cancer immunotherapy. Therefore, the effect of OX40 agonist treatment on T cell function *in vitro* was examined.

[0403] Isolated CD4+ T cells were isolated from PBMCs, stimulated to induce OX40 expression, and then re-stimulated with anti-CD3 in the presence of the agonist anti-human OX40 mAb, as described above. **FIGS. 1A & 1B** demonstrate that T cell proliferation (**FIG. 1A**) and IFNγ production (**FIG. 1B**) were both enhanced by treatment with anti-OX40, as compared to a control treatment.

[0404] In addition, anti-OX40 treatment was found to reduce Treg activity in the *in vitro* Treg suppression assay described above (**FIG. 2**). These results demonstrate that OX40 agonist treatment is able to modulate several critical T cell functions.

[0405] OX40 agonist treatment was next analyzed in several syngeneic mouse tumor models to examine whether OX40 agonism has an effect on tumor growth *in vivo*. Mice were inoculated with various syngeneic tumor cell types to develop tumors, and once these tumors reached a threshold size, mice were treated with murine anti-mouse OX40 mAb or control antibody, as described above.

[0406] As shown in **FIGS. 3A-3C**, anti-OX40 agonist treatment caused a dramatic reduction and durable regression in several tumor models, including EMT6 breast (**FIG. 3A**), Cloudman melanoma (**FIG. 3B**), and CT26 colorectal cancer (CRC) (**FIG. 3C**) tumor models. In contrast, other mouse tumor models showed less responsiveness to treatment, such as the MC38 CRC (**FIG. 4A**), 51Blim10 CRC (**FIG. 4B**), and JC breast (**FIG. 4C**) tumor models. These results demonstrate differential anti-tumor activity of OX40 agonist treatment in mouse tumor models. However, OX40 agonist treatment led to anti-tumor activity, including durable tumor regression, in several tumor models.

Example 2: Baseline immune function associated with gene expression in tumors may be predictive of differential anti-tumor activity of OX40 agonist treatment

[0407] Because of the observation that OX40 agonist treatment leads to differential responsiveness in various tumor models, the expression of more than 90 immune-related

genes was next examined, comparing gene expression in tumor models that were found to be more responsive to OX40 agonist treatment with gene expression in tumor models that were found to be less responsive. As described above, these samples were isolated prior to any treatment with OX40 agonist, when tumors reached a threshold size.

[0408] As shown in **FIG. 5**, higher expression of certain immune activation and Th1 markers may be associated with better responsiveness in EMT6 and CT26 tumors. For example, higher expression of CD8, IFNg, GZMB, PRF1, and PDCA1 may be associated with better responsiveness to anti-OX40 treatment. In addition, higher expression of CXCL9, PTPRC, IL7R, KLRK1, and CXCL1 may also be associated with better responsiveness to anti-OX40 treatment.

[0409] In contrast, higher expression of CCL2, GATA3, IL13, VTCN1, and CSF2 may be associated with poor responsiveness to an anti-OX40 treatment, but lower expression of these genes may be associated with better responsiveness to an anti-OX40 treatment. Genes that are differentially expressed between responsive and non-responsive tumor types are listed below in Table 7. As shown in Table 7, genes that were found to correlate with better responsiveness when expressed at a higher level are classified as positive predictors, whereas genes that were found to correlate with poor responsiveness when expressed at a higher level are classified as negative predictors. These negative predictors may correlate with better responsiveness when expressed at a lower level in a tumor.

Table 7. Differentially expressed genes for predictive biomarker evaluation.

Gene	Predictor
CSF2	Negative
CCL22	Negative
GATA3	Negative
CD8a	Positive
CD8b	Positive
H2-d	Positive
CTLA4	Positive
CD64	Positive
CXCL9	Positive
IFNg	Positive
IDO1	Positive
GZMA	Positive
GZMB	Positive
PRF1	Positive

PDCA1	Positive
KLRK1	Positive
PTPRC	Positive
CXCL1	Positive
ITGAM	Positive
IL7R	Positive
IL13	Negative
EPCAM	Negative
VTCN1	Negative

[0410] In summary, these results indicate that the expression level of specific genes may be associated with responsiveness to OX40 agonist treatment.

Example 3: OX40 agonist treatment induces immune modulation in different tumor models

[0411] In addition to gene expression, other parameters of immune modulation were next examined in various tumor models in response to the OX40 agonist treatment.

[0412] **FIGS. 6A & 6B** show the dose-dependent effects of anti-OX40 treatment on peripheral blood cells in the EMT6 tumor model. Anti-OX40 treatment using murine anti-mouse OX40 mAb led to a dose-dependent reduction in peripheral Treg cells (**FIG. 6A**) and a dose-dependent increase in peripheral CD8 T cell proliferation (**FIG. 6B**).

[0413] T cell sub-populations in tumors were also analyzed. Anti-OX40 treatment caused a reduction in Treg cells in the EMT6 breast tumor model, which showed better responsiveness to the anti-OX40 treatment (**FIG. 7A**). Importantly, the anti-OX40 treatment induced a sustained increase in CD8 tumor infiltrate in the EMT6 model (**FIG. 7B**). In the JC breast tumor model, which was less responsive to the anti-OX40 treatment, the anti-OX40 antibody also led to a reduction in Treg cells in JC tumors (**FIG. 8A**). However, CD8 T cells in JC tumors showed a more slight increase than in EMT6 cells (**FIG. 8B**).

[0414] Gene expression of specific markers of immune activation was also examined in these tumor models. As shown in **FIGS. 9A-9D**, expression of several gene markers was associated with pharmacodynamic (PD) activity in both tumor models, with slightly increased activity in the EMT6 model compared to the JC model. These gene markers included IFNg (**FIG. 9A**), granzyme A (**FIG. 9B**), perforin (**FIG. 9C**), and TNFa (**FIG. 9D**). Table 8 lists the markers for PD activity identified in these experiments.

Table 8. Markers of PD activity

ARG1	ICAM1
CCL2	IDO1
CCL22	IFNg
CCL5	IL10
CCR5	IL12A (TDO2)
CD226	IL13
CD27	IL2
CD274	IL7R
CD28	ITGAM
CD3E	KLRK1
CD40	LAG3
CD8A	MAP4K1
CD8b	MS4A1
CXCL10	PDCD1
CXCL9	PDCD1LG2
EOMES	PRF1
FasL	PTPRC
Fcgr1/CD64	TNF
FOXP3	TNFRSF14
GZMA	TNFRSF9
GZMB	TNFSF4
HAVCR2	

[0415] These results suggest that genes associated with immune activation are induced by anti-OX40 in both EMT6 and JC tumors, but this increase in expression may not reach a threshold required for anti-tumor activity in JC tumors. Importantly, these results identify genes that could be used as markers for PD activity in both responsive and non-responsive tumors.

[0416] Other genes associated with antigen presentation, co-stimulation, and IFNg response were differentially upregulated in EMT6 tumors (**FIGS. 10A-10D**). These gene markers included H2-aa (**FIG. 10A**), CD86 (**FIG. 10B**), ICOS (**FIG. 10C**), and CXCR3 (**FIG. 10D**). Table 9 lists the markers for responsiveness to OX40 agonist treatment identified in these experiments.

Table 9. Markers for responsiveness

BTLA	Fcgr2b/CD32

CD4	Fcgr3/CD16
CD69	H2-aa
CD80	H2-d
CD83	H2-k
CD86	ICOS
CSF2	IL10
CTLA4	PDCA1
CXCR3	TNFRSF18

[0417] These results indicate that enhanced PD modulation by anti-OX40 (murine anti-mouse OX40 mAb) in EMT6 tumors compared to JC tumors may be associated with a better anti-tumor response. Further, these results identify genes that could be used as markers for responsiveness to OX40 agonist treatment.

[0418] In summary, these results demonstrate that OX40 agonists induce potent T cell activation and promote anti-tumor immunity and efficacy in preclinical systems. In addition, the biomarkers identified in these studies may be utilized to confirm mechanism of action, inform dose finding, and guide patient and indication selection in clinical trials.

[0419] All patents, patent applications, documents, and articles cited herein are herein incorporated by reference in their entireties.

CLAIMS

What is claimed is:

1. A method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and

(b) classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to the OX40 agonist treatment.

2. Use of an OX40 agonist in the manufacture of a medicament for a method for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein said method comprises:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R; and

(b) wherein the OX40 agonist is to be administered to the subject if the expression level of said one or more marker genes in the tumor sample obtained from the subject is higher than a reference.

3. Use of an OX40 agonist in the manufacture of a medicament for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, wherein the OX40 agonist is to be administered to the subject, and wherein a tumor sample comprising leukocytes obtained from the subject has been detected to possess increased expression of one or more marker genes are selected from the group consisting of CD8a, CD8b, HLA-B, CTLA4, CD64, CXCL9, IFNg, IDO1, GZMA, GZMB, PRF1, PDCA1, KLRK1, PTPRC, CXCL1, ITGAM, and IL7R, as compared with a reference.

4. The method or use of any one of claims 1-3, wherein said one or more marker genes are selected from the group consisting of CD8a, CD8b, IFNg, GZMA, GZMB, PRF1, and PDCA1.

5. The method or use of any one of claims 1-3, wherein said one or more marker genes are selected from the group consisting of HLA-B, CTLA4, CXCL9, PTPRC, IL7R, KLRK1, and CXCL1.

6. The method or use of any one of claims 1-3, wherein said one or more marker genes are selected from the group consisting of CD64, IDO1, and ITGAM.

7. A method for predicting responsiveness of a subject having cancer to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and

(b) classifying the subject as a responsive or non-responsive subject based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein a decreased expression level of the one or more marker genes as compared with the reference indicates the subject may be responsive to an OX40 agonist treatment.

8. Use of an OX40 agonist in the manufacture of a medicament for a method for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein said method comprises:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein said one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1; and

(b) wherein the OX40 agonist is to be administered to the subject if the expression level of said one or more marker genes in the tumor sample obtained from the subject is lower than a reference.

9. Use of an OX40 agonist in the manufacture of a medicament for treating or delaying progression of cancer in a subject, wherein the OX40 agonist is an anti-OX40 antibody, and wherein the OX40 agonist is to be administered to the subject, and wherein a tumor sample comprising leukocytes obtained from the subject has been detected to possess decreased expression of one or more marker genes are selected from the group consisting of CSF2, CCL22, EPCAM, GATA3, IL13, and VTCN1, as compared with a reference.

10. A method for monitoring pharmacodynamic activity of an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an anti-OX40 agonist antibody, wherein the subject has cancer, and wherein said one or more marker genes are selected from the group consisting of ARG1, CCL2, CCL22, CCL5, CCR5, CD226, CD27, CD274, CD28, CD3E, CD40, CD8A, CD8b, CXCL10, CXCL9, EOMES, FasL, Fcgr1/CD64, FOXP3, GZMA, GZMB, HAVCR2, ICAM1, IDO1, IFNg, IL10, IL12A (TDO2), IL13, IL2, IL7R, ITGAM, KLRK1, LAG3, MAP4K1, MS4A1, PDCD1, PDCD1LG2, PRF1, PTPRC, TNF, TNFRSF14, TNFRSF9, and TNFSF4; and

(b) determining the treatment as demonstrating pharmacodynamic activity based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates pharmacodynamic activity to the OX40 agonist treatment.

11. The method of claim 10, wherein said one or more marker genes are selected from the group consisting of CD3, CD8, IFNg, GZMA, GZMB, PRF1, TNFa, PDCD1, and CD274.

12. A method for monitoring responsiveness of a subject to an OX40 agonist treatment, wherein the OX40 agonist is an anti-OX40 antibody, said method comprising:

(a) measuring the expression level of one or more marker genes in a tumor sample comprising leukocytes obtained from the subject, wherein the subject has been treated with an anti-OX40 agonist antibody, wherein the subject has cancer, and wherein said one or more marker genes are selected from the group consisting of BTLA, CD4, CD69, CD80, CD83, CD86, CSF2, CTLA4, CXCR3, Fcgr2b/CD32, Fcgr3/CD16, HLA-DQA1, HLA-B, HLA-A, ICOS, IL10, PDCA1, and TNFRSF18; and

(b) classifying the subject as responsive or non-responsive to said treatment based on the expression level of said one or more marker genes in the tumor sample obtained from the subject, as compared with a reference, wherein an increased expression level of the one or more marker genes as compared with the reference indicates a responsive subject.

13. The method of claim 12, wherein said one or more marker genes are selected from the group consisting of CD80, CD86, ICOS, HLA-DQA1, and CXCR3.

14. The method of claim 12 or claim 13, wherein responsiveness comprises immune activation and/or treatment efficacy.

15. The method or use of any one of claims 1-14, wherein the expression level of said one or more marker genes is normalized to the expression level of a reference gene in the sample.

16. The method or use of claim 15, wherein the reference gene is a housekeeping gene.

17. The method or use of any one of claims 1-16, wherein the expression level of said one or more marker genes is mRNA expression level.

18. The method or use of claim 17, wherein the mRNA expression level is measured by an assay selected from the group consisting of quantitative PCR, semi-quantitative PCR, nucleotide microarray, RNA-seq, in situ hybridization, and Northern blotting.

19. The method or use of any one of claims 1-16, wherein the expression level of said one or more marker genes is protein expression level.

20. The method or use of claim 19, wherein the protein expression level is measured by Western blotting, peptide microarray, immunohistochemistry, flow cytometry, or mass spectrometry.

21. The method or use of any one of claims 1-20, wherein the cancer is selected from the group consisting of colorectal cancer, non-small cell lung cancer, renal cell carcinoma, bladder cancer, ovarian cancer, glioblastoma, neuroblastoma, melanoma, breast carcinoma, gastric cancer, and hepatocellular carcinoma.

22. The method or use of claim 21, wherein the breast carcinoma is triple-negative breast carcinoma.

23. The method or use of any one of claims 1-22, wherein the antibody is a monoclonal antibody.

24. The method or use of any one of claims 1-22, wherein the antibody is a humanized or human antibody.

25. The method or use of any one of claims 1-22, wherein the antibody comprises an IgG1 Fc region.

26. The method or use of any one of claims 1-22, wherein the antibody comprises an IgG4 Fc region.

27. The method or use of any one of claims 1-22, wherein the antibody comprises an Fc region comprising a mutation that decreases binding to an Fc receptor.

28. The method or use of any one of claims 1-22, wherein the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2, 8 or 9; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3, 10, 11, 12, 13, or 14; (c) HVR-H3 comprising the amino acid

sequence of SEQ ID NO:4, 15 or 19; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:7, 22, 23, 24, 25, 26, 27 or 28.

29. The method or use of any one of claims 1-22, wherein the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:7.

30. The method or use of any one of claims 1-22, wherein the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:26.

31. The method or use of any one of claims 1-22, wherein the antibody comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:2; (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:3; (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:4; (d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:5; (e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:6; and (f) HVR-L3 comprising an amino acid sequence selected from SEQ ID NO:27.

32. The method or use of any one of claims 1-22, wherein the antibody is MEDI6469 or MEDI0562.