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(54) Title: METHODS AND COMPOSITIONS FOR TREATMENT OF CANCER

(57) Abstract: The present invention relates to VEGF-binding agents, DLL4-binding agents, VEGF/DLL4 bispecific binding agents, and methods of using the agents for treating diseases such as cancer, particularly pancreatic, colorectal, and endometrial cancers. Also provided are methods, compositions, and kits for treatment of tumors or cancer using combinations that include a VEGF/DLL4 bispecific agent and one or more chemotherapeutic agents (e.g., gemcitabine and ABRAXANE®; leucovorin, 5- fluorouracil, and irinotecan; and paclitaxel and carboplatin). The present invention further provides methods of using the agents or combinations of agents to inhibit growth of a pancreatic, colorectal, or endometrial tumor. Also described are methods of treating cancer, particularly pancreatic, colorectal, and endometrial cancer, comprising administering a therapeutically effect amount of an agent, antibody, or therapeutic combination of the present invention to a patient having a tumor or cancer.

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METHODS AND COMPOSITIONS FOR TREATMENT OF CANCER

CROSS-REFERENCE TO RELATED APPLICATONS

[0001] This application claims the priority benefit of U.S. Provisional Application No. 62/222,505, filed September 23, 2015, which is hereby incorporated by reference herein in its entirety.

FIELD OF THE INVENTION

[0002] The present invention generally relates to methods for treating cancer, particularly colorectal, ovarian, pancreatic, and endometrial cancer, using antibodies and other agents that bind VEGF, DLL4, or both VEGF and DLL4, particularly anti-VEGF/anti-DLL4 bispecific antibodies, optionally in combination with additional therapeutic agents. The invention also relates to compositions and kits including the combinations.

BACKGROUND OF THE INVENTION

[0003] Colorectal cancers are one of the most common types of cancer in the United States. More than 132,000 people are diagnosed with colon cancer each year as of 2015, according to the National Cancer Institute. Approximately 1 in 19 people, or a little more than 5% of Americans, will develop colon or rectal cancer in their lifetimes.

[0004] Approximately 1.3% of women will be diagnosed with ovarian cancer in their lifetimes, according to 2010-2012 data from the National Cancer institute. In 2016, the National Cancer Institute estimates there will be over 60,000 new cases of ovarian cancer and over 24,000 ovarian cancer deaths.

[0005] Pancreatic cancers, while only making up 2% of all cancer diagnoses, are the fifth leading cause of cancer deaths in the United States. More than 48,000 people are diagnosed with, and more than 40,000 people die from pancreatic cancer each year as of 2015, according to the National Cancer Institute.

[0006] Endometrial cancer, another common type of cancer, arises from the uterine lining. More than 54,000 new cases of endometrial cancer diagnosed each year as of 2015 in the United States, according to the National Cancer Institute. Endometrial cancer is the most common gynecologic malignancy in the United States and accounts for 6% of all cancers in women, according to the National Cancer Institute.

[0007] The focus of cancer drug research is shifting toward targeted therapies aimed at genes, proteins, and pathways involved in human cancer. There is a need for new agents targeting signaling pathways and new combinations of agents that target multiple pathways that could provide therapeutic benefit for cancer patients. Thus, biomolecules (e.g., bispecific antibodies) that disrupt multiple signaling pathways are a potential source of new therapeutic agents for cancer.

[0008] Signaling pathways normally connect extracellular signals to the nucleus leading to expression of genes that directly or indirectly control cell growth, differentiation, survival and death. In melanoma as

well as a wide variety of cancers, signaling pathways are dysregulated and may be linked to tumor initiation and/or progression. Signaling pathways implicated in human oncogenesis include, but are not limited to, the Notch pathway, the VEGF pathway, the Ras-Raf-MEK-ERK or MAPK pathway, the PI3K-AKT pathway, the CDKN2A/CDK4 pathway, the Bcl-2/TP53 pathway, and the Wnt pathway.

[0009] Angiogenesis plays an important role in the pathogenesis of a number of disorders, including solid tumors and metastasis. The production of new blood vessels is essential for providing oxygen and nutrients for the growth and spread of a tumor, and therefore angiogenesis is a good target for cancer therapeutics.

[0010] Angiogenesis involves a family of proteins acting as angiogenic activators, including vascular endothelial growth factor (VEGF-A), VEGF-B, VEGF-C, VEGF-E, and their respective receptors (VEGFR-1, VEGFR-2, and VEGFR-3). VEGF-A, also referred to as VEGF or vascular permeability factor (VPF), exists in several isoforms that arise from alternative splicing of mRNA of a single VEGF gene, with $VEGF_{165}$ being the most biologically relevant isoform.

[0011] Anti-VEGF antibodies have been shown to suppress the growth of tumor cells *in vitro* and *in vivo*. A humanized anti-VEGF monoclonal antibody, bevacizumab (AVASTIN) has been developed and approved in the United States as a cancer therapeutic.

[0012] The Notch signaling pathway is a universally conserved signal transduction system. It is involved in cell fate determination during development including embryonic pattern formation and post-embryonic tissue maintenance. In addition, Notch signaling has been identified as a critical factor in the maintenance of hematopoietic stem cells.

[0013] The Notch pathway has been linked to the pathogenesis of both hematologic and solid tumors and cancers. Numerous cellular functions and microenvironmental cues associated with tumorigenesis have been shown to be modulated by Notch pathway signaling, including cell proliferation, apoptosis, adhesion, and angiogenesis (Leong et al., 2006, *Blood*, 107:2223-2233). In addition, Notch receptors and/or Notch ligands have been shown to play potential oncogenic roles in a number of human cancers, including acute myelogenous leukemia, B cell chronic lymphocytic leukemia, Hodgkin lymphoma, multiple myeloma, T-cell acute lymphoblastic leukemia, brain cancer, breast cancer, cervical cancer, colon cancer, lung cancer, pancreatic cancer, prostate cancer, and skin cancer (Leong et al., 2006, *Blood*, 107:2223-2233).

[0014] Delta-like 4 ligand (DLL4) is an important component of the Notch pathway and has been identified as a target for cancer therapy. DLL4 is a Notch ligand, characterized by an N-terminal domain, a Delta/Serrate/Lag-2 (DSL) domain and tandem EGF-like repeats within the extracellular domain. It has been reported that DLL4 is induced by VEGF and that DLL4 may act as a negative feedback regulator for vascular proliferation.

[0015] Anti-DLL4 antibodies have been shown to enhance angiogenic sprouting and branching which leads to non-productive angiogenesis and decreased tumor growth (Noguera-Troise et al., 2006, *Nature*, 444:1032-1037). In addition, an anti-DLL4 antibody, demcizumab (also known as OMP-21M18 or 21M18), has been shown to inhibit tumor growth and reduce the frequency of cancer stem cells in xenograft tumor models (Hoey et al., 2009, *Cell Stem Cell*, 5:168-177; U.S. Patent No. 7,750,124).

[0016] Although there have been significant strides in development of monoclonal antibodies for use in cancer treatments, there is still great potential for further improvements. One class of antibody molecules with the promise of enhanced potency and/or reduced side effects (e.g., toxicity) is bispecific antibodies.

[0017] Early bispecific molecules were mainly generated using chemical cross-linking of two antibodies, or were hybrid hybridomas or "quadromas". One success of the quadroma format is triomabs, which are mouse/rat combinations that demonstrate a preferential species-specific heavy/light chain pairing. More recently, advances in antibody engineering have provided a wide variety of new antibody formats, including, but not limited to, tandem scFv (bi-scFv), diabodies, tandem diabodies (tetra-bodies), single chain diabodies, and dual variable domain antibodies.

[0018] It is one of the objectives of the present invention to provide improved cancer treatment, particularly using bispecific antibodies that specifically bind human VEGF and human DLL4 optionally in combination with other anti-cancer agent(s), to treat cancer, particularly colorectal cancer, ovarian cancer, pancreatic cancer, and endometrial cancer.

SUMMARY OF THE INVENTION

[0019] The present invention provides methods for treatment of cancer, for example, colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial cancer, using antibodies or other binding agents that bind VEGF, DLL4, or both VEGF and DLL4, optionally in combination with additional anti-cancer therapeutics (e.g., any of those described herein). The invention also features compositions and kits that include therapeutic combinations.

[0020] In a first aspect, the invention provides methods of inhibiting growth of a tumor, comprising contacting the tumor with an effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the tumor is a colorectal tumor. In certain embodiments the contacting takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination is used as a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0021] In another aspect, the invention provides a method of inhibiting the growth of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or

other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the tumor is a colorectal tumor. In certain embodiments the administering takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination is used as a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0022] In another aspect, the invention provides a method of reducing the tumorigenicity of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the tumor is a colorectal tumor. In certain embodiments the contacting takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination is used as a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0023] In another aspect, the invention provides a method of reducing the frequency of cancer stem cells in a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the tumor is a colorectal tumor. In certain embodiments the administering takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination is used as a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0024] In other aspects, the invention provides methods of treating cancer in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the tumor is a colorectal tumor. In certain embodiments the administering takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination is used as a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0025] In another aspect, the invention provides a method of modulating angiogenesis in a subject who has cancer, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with leucovorin, 5-fluorouracil, and irinotecan. In certain embodiments, the cancer is colorectal cancer. In certain embodiments the administering takes place after the tumor has failed to respond to another anti-cancer treatment (e.g., the combination used as

a second-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0026] In certain embodiments where the method includes administration of leucovorin, 5-fluoroufacil, and irinotecan, these agents are administered according to the FOLFIRI protocol.

[0027] In other aspects, the invention provides methods of inhibiting growth of a tumor, comprising contacting the tumor with an effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the contacting takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0028] In another aspect, the invention provides a method of inhibiting growth of an ovarian, primary peritoneal, or fallopian tumor (e.g., a platinum resistant tumor) comprising contacting the tumor or tumor cells with an effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0029] In another aspect, the invention provides a method of inhibiting growth of an ovarian, primary peritoneal, or fallopian tumor (e.g., a platinum resistant tumor) in a subject comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0030] In another aspect, the invention provides a method of reducing the tumorigenicity of a ovarian, primary peritoneal, or fallopian tumor (e.g., a platinum resistant tumor) in a subject comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0031] In another aspect, the invention provides a method of reducing the frequency of cancer stem cells in an ovarian, primary peritoneal, or fallopian tumor (e.g., a platinum resistant tumor) in a subject comprising administering to the subject a therapeutically effective amount of an antibody (or other

binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0032] In another aspect, the invention provides a method of treating ovarian, primary peritoneal, or fallopian cancer (e.g., a platinum resistant cancer) in a subject comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0033] In another aspect, the invention provides a method of modulating angiogenesis in a subject that has ovarian, primary peritoneal, or fallopian cancer (e.g., a platinum resistant cancer) comprising administering to the subject a therapeutically effective amount an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. In some embodiments, the antibody or binding agent is administered in combination with a taxane (e.g., paclitaxel). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0034] In any of the above six aspects, the antibody, other binding agent, or combination including the antibody or binding agent is administered following failure of at least one, two, three, or four prior therapies (e.g., failure of more than two, such as three or four, prior therapies) and/or have received a prior anti-VEGF agent (e.g., an anti-VEGF antibody such as bevacizumab).

[0035] In another aspect, the invention provides methods of inhibiting growth of a tumor, comprising contacting the tumor with an effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and nab-paclitaxel (ABRAXANE®). In certain embodiments, the tumor is a pancreatic tumor. In certain embodiments, the contacting takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0036] In another aspect, the invention provides a method of inhibiting the growth of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and nab-paclitaxel (ABRAXANE®). In certain embodiments, the tumor is a pancreatic tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is

used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0037] In another aspect, the invention provides a method of reducing the tumorigenicity of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and ABRAXANE®. In certain embodiments, the tumor is a pancreatic tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0038] In another aspect, the invention provides a method of reducing the frequency of cancer stem cells in a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and ABRAXANE®. In certain embodiments, the tumor is a pancreatic tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0039] In other aspects, the invention provides methods of treating cancer in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and ABRAXANE®. In certain embodiments, the cancer is pancreatic cancer. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0040] In another aspect, the invention provides a method of modulating angiogenesis in a subject who has cancer, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with gemcitabine and ABRAXANE® described herein. In certain embodiments, the cancer is pancreatic cancer. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0041] In another aspect, the invention provides a method of inhibiting the growth of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or

other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0042] In another aspect, the invention provides a method of reducing the tumorigenicity of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0043] In another aspect, the invention provides a method of reducing the frequency of cancer stem cells in a tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0044] In other aspects, the invention provides methods of treating cancer in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0045] In another aspect, the invention provides a method of modulating angiogenesis in a subject who has cancer, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein, in combination with paclitaxel and carboplatin. In certain embodiments, the cancer is endometrial cancer. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the combination is used as a first-line

treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0046] In other aspects, the invention provides methods of inhibiting growth of an endometrial tumor, comprising contacting the tumor with an effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the contacting is performed in combination with paclitaxel and carboplatin. In certain embodiments, the contacting takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0047] In another aspect, the invention provides a method of inhibiting the growth of an endometrial tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the administering is performed in combination with paclitaxel and carboplatin. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0048] In another aspect, the invention provides a method of reducing the tumorigenicity of an endometrial tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the administering is in combination with paclitaxel and carboplatin. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0049] In another aspect, the invention provides a method of reducing the frequency of cancer stem cells in an endometrial tumor in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the administering is in combination with paclitaxel and carboplatin. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0050] In other aspects, the invention provides methods of treating endometrial cancer in a subject, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the administering is in combination with paclitaxel and carboplatin. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0051] In another aspect, the invention provides a method of modulating angiogenesis in a subject who has endometrial cancer, comprising administering to the subject a therapeutically effective amount of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In certain embodiments, the administering is in combination with paclitaxel and carboplatin. In certain embodiments, the administering takes place prior to treatment with another anti-cancer treatment (e.g., the antibody or combination including the antibody is used as a first-line treatment). In certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0052] In another aspect, the invention features a method of managing blood pressure in a subject that is indicated for or receiving treatment with an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies or other binding agents described herein. The method comprises (a) determining the blood pressure in the subject, wherein blood pressure greater than 140/90 is indicative of hypertension; (b) administering to the hypertensive subject hydralazine or clonidine if the subject's systolic pressure exceeds 180 mm Hg for acute (e.g., no longer than 48-72 hours) blood pressure reduction; (c) administering to the hypertensive subject an initial dose of one of amlodipine and Procardia XL® (e.g., 5 mg orally daily for amlodipine or 30-60 mg orally daily for Procardia XL®) for chronic blood pressure management; (d) adjusting the dose of the amlodipine or Procardia XL®, if blood pressure is not adequately controlled (e.g., reduced to under 140/90) by the initial dose, up to a maximum dose (e.g., 10 mg orally daily for amlodipine or 120 mg orally daily for Procardia XL®); (e) administering a second antihypertensive medication, if the blood pressure is not adequately controlled (e.g., reduced to under 140/90) by the maximum dose of amlodipine or Procardia XL®, wherein the second antihypertensive medication is an angiotensin-converting-enzyme (ACE) inhibitor or a beta blocker; (f) administering a third antihypertensive medication to the subject, if the blood pressure is not adequately controlled by the first two medications, wherein the third antihypertensive medication is an ACE inhibitor or beta-blocker, whichever was not used as the second antihypertensive medication; and (g) administering to the subject a dose of an antibody (or other binding agent) that binds VEGF, DLL4, or both VEGF and DLL4, including any of the antibodies (or other binding agents) described herein. In

certain embodiments, the antibody is an anti-DLL4/VEGF bispecific, e.g., any described herein such as 305B83.

[0053] In any of the aspects described above or elsewhere herein, the subject is a human.

[0054] The binding agents (e.g., antibodies) used in the present invention can bind VEGF, DLL4, or both VEGF and DLL4 (VEGF/DLL4-binding agents). Agents that bind VEGF or DLL4, as well as at least one additional antigen or target, and pharmaceutical compositions of such agents, can also be used. In certain embodiments, the binding agents are polypeptides, such as antibodies, antibody fragments, and other polypeptides related to such antibodies. In certain embodiments, the binding agents are antibodies that specifically bind human VEGF. In some embodiments, the binding agents are antibodies that specifically bind human DLL4. In some embodiments, the binding agents are bispecific antibodies that specifically bind human VEGF and human DLL4. Also provided are compositions, such as pharmaceutical compositions, and kits that include a binding agent described herein. These compositions and kits can be used in any of the methods described herein.

[0055] In some embodiments, the binding agent inhibits binding of VEGF to at least one VEGF receptor. In some embodiments, the binding agent inhibits binding of VEGF to VEGFR-1 and/or VEGFR-2. In some embodiments, the binding agent modulates angiogenesis. In certain embodiments, the antibody or other binding agent further specifically binds to and/or inhibits human DLL4 in addition to human VEGF.

[0056] In some embodiments, the binding agent is an antibody which comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0057] In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region having at least 80% sequence identity to SEQ ID NO:11; and/or a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the binding agent comprises a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:11; and/or a light chain variable region having at least 90% sequence identity to SEQ ID NO:12. In certain embodiments, the binding agent comprises a heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11; and/or a light chain variable region having at least 95% sequence identity to SEQ ID NO:12. In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region of SEQ ID NO:11; and/or a light chain variable region of SEQ ID NO:12.

[0058] In some embodiments, the binding agent is antibody 219R45, bispecific antibody 219R45-MB-21M18 (also known as 305B18), bispecific antibody 219R45-MB-21R79 (also known as 305B79), bispecific antibody 219R45-MB-21R75 (also known as 305B75), or bispecific antibody 219R45-MB-21R83 (also known as 305B83).

[0059] In another aspect, the invention provides a binding agent, such as an antibody, that specifically binds human DLL4. In some embodiments, the binding agent inhibits binding of DLL4 to at least one Notch receptor. In some embodiments, the binding agent inhibits binding of DLL4 to Notch1, Notch2, Notch3, and/or Notch4. In some embodiments, the binding agent inhibits Notch signaling. In some embodiments, the binding agent promotes unproductive angiogenesis. In certain embodiments, the antibody or other binding agent further specifically binds to and/or inhibits human VEGF in addition to human DLL4.

[0060] In some embodiments, the binding agent is an antibody that binds human DLL4 and comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the antibody comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0061] In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:10; and/or a light chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:12. In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region of SEQ ID NO:10; and a light chain variable region of SEQ ID NO:12.

[0062] In some embodiments, the binding agent is antibody 21R79 or bispecific antibody 219R45-MB-21R79 (305B79).

[0063] In some embodiments, the binding agent is an antibody which comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0064] In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:58; and/or a light chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:12. In certain

embodiments, the binding agent is an antibody that comprises a heavy chain variable region of SEQ ID NO:58; and a light chain variable region of SEQ ID NO:12.

[0065] In some embodiments, the binding agent is antibody 21R75 or bispecific antibody 219R45-MB-21R75 (305B75).

[0066] In some embodiments, the binding agent is an antibody which comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0067] In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:64; and/or a light chain variable region having at least 90% or at least 95% sequence identity to SEQ ID NO:12. In certain embodiments, the binding agent is an antibody that comprises a heavy chain variable region of SEQ ID NO:64; and a light chain variable region of SEQ ID NO:12.

[0068] In some embodiments, the binding agent is antibody 21R83 or bispecific antibody 219R45-MB-21R83 (305B83).

[0069] In certain embodiments of each of the aforementioned aspects or embodiments, as well as other aspects and/or embodiments described elsewhere herein, the binding agent is a bispecific antibody. In some embodiments, the bispecific antibody specifically binds human VEGF and a second target. In some embodiments, the bispecific antibody specifically binds human DLL4 and a second target. In some embodiments, the bispecific antibody specifically binds both human VEGF and human DLL4. In some embodiments, the bispecific antibody modulates angiogenesis. In certain embodiments, the bispecific antibody inhibits Notch signaling. In some embodiments, the bispecific antibody modulates angiogenesis and inhibits Notch signaling. In some embodiments, the bispecific antibody reduces the number or frequency of cancer stem cells. In certain embodiments, the bispecific antibody comprises two identical light chains. In certain embodiments the bispecific antibody is an IgG antibody (e.g., IgG2 antibody).

[0070] In some embodiments, the bispecific antibody comprises: a first antigen-binding site that specifically binds human VEGF, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19). In some embodiments, the bispecific antibody further comprises: a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises: a first antigen-binding site that specifically binds human VEGF, wherein

the first antigen-binding site comprises (a) a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0071] In certain embodiments, the bispecific antibody comprises: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody further comprises: a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0072] In some embodiments, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a

heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISSYNGATNYNQKFKG (SEQ ID NO:15), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the a bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some

embodiments, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[0073] In some embodiments, the bispecific antibody that specifically binds human VEGF, and comprises: a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:11, and/or a light chain variable region having at least 90% sequence identity to SEQ ID NO:12. In some embodiments, the bispecific antibody specifically binds human VEGF, and comprises a heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11 and/or a light chain variable region having at least 95% sequence identity to SEQ ID NO:12.

[0074] In some embodiments, the bispecific antibody specifically binds human DLL4, and comprises a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64; and/or a light chain variable region having at least 90% sequence identity to SEQ ID NO:12. In some embodiments, the bispecific antibody specifically binds human DLL4, and comprises a heavy chain variable region having at least 95% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64; and/or a light chain variable region having at least 95% sequence identity to SEQ ID NO:12.

[0075] In some embodiments, the bispecific antibody specifically binds human VEGF and human DLL4, and comprises: (a) a first heavy chain variable region having at least 90% sequence identity to SEQ ID NO:11; (b) a second heavy chain variable region having at least 90% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64; and (c) a first and a second light chain variable region having at least 90% sequence identity to SEQ ID NO:12. In some embodiments, the VEGF/DLL4 bispecific antibody comprises (a) a first heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11; (b) a second heavy chain variable region having at least 95% sequence identity to SEQ ID NO:9; and (c) a first and a second light chain variable region having at least 95% sequence identity to SEQ ID NO:12. In some embodiments, the VEGF/DLL4 bispecific antibody comprises (a) a first heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11; (b) a second heavy chain variable region having at least 95% sequence identity to SEQ ID NO:10; and (c) a first and a second light chain variable region having at least 95% sequence identity to SEQ ID NO:12. In some embodiments, the

VEGF/DLL4 bispecific antibody comprises (a) a first heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11; (b) a second heavy chain variable region having at least 95% sequence identity to SEQ ID NO:58; and (c) a first and a second light chain variable region having at least 95% sequence identity to SEQ ID NO:12. In some embodiments, the VEGF/DLL4 bispecific antibody comprises (a) a first heavy chain variable region having at least 95% sequence identity to SEQ ID NO:11; (b) a second heavy chain variable region having at least 95% sequence identity to SEQ ID NO:64; and (c) a first and a second light chain variable region having at least 95% sequence identity to SEQ ID NO:12.

[0076] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising (a) a first antigen-binding site that binds human VEGF with a K_D between about 0.1nM and about 1.0nM and (b) a second antigen-binding site that specifically binds human DLL4 with a K_D between about 0.1nM and about 20nM. In certain embodiments, the bispecific antibody comprises two identical light chains.

[0077] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody selected from the group consisting of 219R45-MB-21M18 (305B18), 219R45-MB-21R79 (305B79), 219R45-MB-21R75 (305B75), and 219R45-MB-21R83 (305B83).

[0078] In certain embodiments of each of the aforementioned aspects, as well as other aspects and/or embodiments described elsewhere herein, the binding agent or antibody is isolated.

[0079] In another aspect, the methods, compositions, or kits of the invention use a polypeptide selected from the group consisting of: SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64. In some embodiments, the polypeptide is isolated. In certain embodiments, the polypeptide is substantially pure. In certain embodiments, the polypeptide is an antibody or part of an antibody, such as an antibody fragment.

[0080] In another aspect, the methods, compositions, and kits of the invention employ an isolated polynucleotide molecule including a polynucleotide that encodes the binding agents and/or polypeptides of each of the aforementioned aspects, as well as other aspects and/or embodiments described herein. In some embodiments, the polynucleotide comprises a sequence selected from the group consisting of: SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:40, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, and SEQ ID NO:74. The invention further provides expression vectors that comprise the polynucleotides, as well as cells that comprise the expression vectors and/or the polynucleotides. In some embodiments, the cell is a prokaryotic cell or a eukaryotic cell.

[0081] In any of the embodiments described herein, the VEGF/DLL4 binding agent (e.g., 305B83) may be administered at a dose between 0.1 mg/kg and 20 mg/kg or between 0.5 mg/kg and 10 mg/kg or about 0.5, 1, 2.5, 3, 4, 5, 10, or 15 mg/kg. In some embodiments, the dose is 3 mg/kg. In other embodiments, the dose is 5 mg/kg. In other embodiments, the dose is 10 mg/kg. In some embodiments, the dose is administered every two weeks (e.g., at 1 mg/kg, 3 mg/kg, 5, mg/kg, 10 mg/kg, or 15 mg/kg). In some embodiments, the dose is administered every three weeks. In other embodiments, the dose is administered every week, every ten days, every four weeks, every six weeks, or every two months.

[0082] In embodiments, involving combinations of a VEGF/DLL4-binding agent and an additional therapeutic(s), the agent and additional therapeutics may be administered in any order or concurrently. In some embodiments, treatment with a VEGF/DLL4-binding agent (e.g., an antibody) can occur prior to, concurrently with, or subsequent to administration of the additional therapeutics. Combined administration may include co-administration, either in a single pharmaceutical formulation or using separate formulations, or consecutive administration in either order but generally within a time period such that all active agents can exert their biological activities simultaneously. Preparation and dosing schedules for such chemotherapeutic agents can be used according to manufacturers' instructions or as determined empirically by the skilled practitioner. Preparation and dosing schedules for such chemotherapy are also described in *The Chemotherapy Source Book, 4th Edition*, 2008, M. C. Perry, Editor, Lippincott, Williams & Wilkins, Philadelphia, PA.

[0083] In certain embodiments, the VEGF/DLL4-binding agent and an additional therapeutic(s) will be administered substantially simultaneously or concurrently. For example, a subject may be given a VEGF/DLL4-binding agent (e.g., an antibody) while undergoing a course of treatment with a second therapeutic agent (e.g., chemotherapy). In certain embodiments, a VEGF/DLL4-binding agent will be administered within 1 year of the treatment with an additional therapeutic agent. In certain alternative embodiments, a VEGF/DLL4-binding agent will be administered within 10, 8, 6, 4, or 2 months of any treatment with an additional therapeutic agent. In certain other embodiments, a VEGF/DLL4-binding agent will be administered within 4, 3, 2, or 1 weeks of any treatment with an additional therapeutic agent. In some embodiments, a VEGF/DLL4-binding agent will be administered within 5, 4, 3, 2, or 1 days of any treatment with an additional therapeutic agent. It will further be appreciated that the two (or more) agents or treatments may be administered to the subject within a matter of hours or minutes (i.e., substantially simultaneously).

[0084] Where aspects or embodiments of the invention are described in terms of a Markush group or other grouping of alternatives, the present invention encompasses not only the entire group listed as a whole, but also each member of the group individually and all possible subgroups of the main group, and also the main group absent one or more of the group members. The present invention also envisages the explicit exclusion of one or more of any of the group members in the claimed invention.

BRIEF DESCRIPTIONS OF THE DRAWINGS

[0085] **Figure 1A** is a schematic diagram showing the escalation and expansion for the phase 1B clinical trials using the combination of an anti-VEGF/DLL4 bispecific antibody (305B83) with paclitaxel (TAXOL) to treat platinum-resistant ovarian, primary peritoneal, or fallopian tube cancer.

[0086] **Figure 1B** is a schematic diagram showing the escalation and expansion for the phase 1B clinical trials using the combination of an anti-VEGF/DLL4 bispecific antibody (305B83) with paclitaxel (TAXOL) and carboplatin ("Carbo") to treat endometrial cancer.

[0087] **Figures 2A** and **2B** are schematic diagrams showing the escalation and expansion for the phase 1B clinical trials using the combination of an anti-VEGF/DLL4 bispecific antibody (305B83) with FOLFORI as a second-line treatment for metastatic colorectal cancer.

[0088] **Figure 3** is a schematic diagram showing the escalation and expansion for the phase 1B clinical trials using the combination of an anti-VEGF/DLL4 bispecific antibody (305B83) with gemcitabine and ABRAXANE® as first-line treatment for metastatic pancreatic cancer.

[0089] **Figure 4** is graph showing anti-tumor activity in ovarian xenograft tumors resulting from simultaneous, complete blockade of DLL4 and VEGF. OMP-OV40 ovarian tumors were treated with either control mAb, bevacizumab+B20 (anti-mVEGF), demcizumab+21R30 (anti-mDLL4), or 305B83 plus 21R30 and B20 at 10 mg/kg once a week for 4 weeks. *:$p<0.05$ vs. control mAb, **: $p<0.05$ vs. anti-VEGF or anti-DLL4 alone by two-way ANOVA.

[0090] **Figures 5A-5C** are photomicrographs and graphs showing anti-VEGF inhibition of angiogenesis is dominant over anti-DLL4 hyperproliferation in animals receiving 305B83. Figure 5A shows expression of CD31, perfusion, and hypoxia in tumors receiving a control mAb, bevacizumab+B20 (anti-mVEGF), demcizumab+21R30 (anti-mDLL4), or 305B83+21R30 and B20. Figures 5B and 5C show expression of murine stromal/vascular genes and human genes, respectively, receiving control mAb, bevacizumab+B20, demcizumab+21R30, or 305B83+(21R30 and B20) from left to right for each gene.

[0091] **Figures 6A** and **6B** are graphs showing comparison of 305B83 with anti-hDLL4 and anti-hVEGF alone at a suboptimal dose. Ovarian tumor OMP-OV40 (Figure 6A) and gastric tumor OMP-STM1 (Figure 6B) were treated with control mAb, 3 mg/kg of demcizumab+21R30 (anti-mDLL4), bevacizumab+B20 (anti-mVEGF) or 305B83 plus 21R30 and B20 once a week for four weeks. *:$p<0.05$ vs. control mAb; **: $p<0.05$ vs. anti-VEGF or anti-DLL4 alone by two-way ANOVA.

[0092] **Figures 7A** and **7B** are graphs showing delay in tumor recurrence by 305B83+21R30 and B20 following chemotherapy termination. Ovarian OMP-OV19 and pancreatic OMP-PN42-tumor bearing animals were randomized and treatment began when mean tumor volumes reached approximately 100–150 mm^3. NOD.SCID mice were treated with 15 mg/kg paclitaxel in OMP-OV19 and 10 mg/kg gemcitabine plus 30 mg/kg nab-pacltiaxel in OMP-PN42 with or without antibody once a week for 4

weeks, followed by a chemotherapy for 3-4 weeks. Thereafter, treatment was discontinued and tumor growth was monitored up to 2 months.

[0093] **Figure 8A** and **8B** are a graph and photographs, respectively, showing that 305B83 inhibits growth of luciferase-labeled OMP-C8 tumors implanted into human skin transplants. Figure 8A shows the effect of 305B18 on OMP-C8 colon xenograft tumor growth implanted into human skin transplants. Neonatal foreskin graft of about 2 cm^2 was implanted into the lateral trunk of anesthetized NOD/SCID mice. Luciferase-labeled human OMP-C8 colon tumor cells were then injected intradermally into the human skin graft 6 weeks post implant. Treatment was initiated 2 weeks later. Tumor growth was monitored by measurement of bioluminescence with the IVIS-200 Imaging System (Caliper Life Sciences). The control mAb, demcizumab, bevacizumab, and 305B18 were given at 25 mg/kg intraperitoneally once a week. **: $p<0.0001$, vs. Control mAb, +: $p<0.01$ vs. demcizumab, *: $p<0.05$ vs. bevacizumab. Figure 8B shows images of tumor size in the control and antibody-treated mice.

[0094] **Figure 9** is a graph showing that DLL4/VEGF bispecific is active in combination with gemcitabine in pancreatic cancer. OMP-PN8 pancreatic tumor cells were injected into NOD-SCID mice. Tumors were allowed to grow for 24 days until they had reached an average tumor volume of 110 mm^3. Tumor-bearing mice were randomized into 4 groups and treated with gemcitabine with either a control antibody, bevacizumab, demcizumab, or anti-DLL4/VEGF bispecific antibody. After four weeks of combination treatment, the gemcitabine was discontinued and the antibody treatments were maintained. Gemcitabine was dosed at 70 mg/kg, weekly. Control Ab, demcizumab, and bevacizumab were dosed 15 mg/kg and the bispecific was dosed 30 mg/kg.

[0095] **Figure 10** is a graph showing reduction of tumor-initiating cell frequency from anti-DLL4 activity. OMP-PN8 pancreatic tumor-bearing mice were treated with either control antibody, demcizumab, bevacizumab, or an anti-DLL4/VEGF bispecific. After four weeks of treatment, tumors were harvested, and the human tumor cells in the xenograft were purified. Ninety tumor cells from each treatment group were injected into new cohorts of 10 mice. Tumors were allowed to grow for 83 days without any further treatment. The volumes of the individual mice in the experiment are shown in the graph.

[0096] **Figure 11** is a graph showing activity of anti-DLL4/VEGF bispecific in combination with gemcitabine plus nab-paclitaxel (ABRAXANE®). OMP-PN42 tumors were implanted in NOD-SCID mice. Tumor-bearing mice (n=10/group) were treated with control Ab, gemcitabine alone (30 mg/kg weekly), gemcitabine plus nab-paclitaxel, or the combination of anti-DLL4/VEGF (305B83, 21R30, B20) and gemcitabine plus nab-paclitaxel. Antibodies were dosed 10 mg/kg, weekly.

DETAILED DESCRIPTION OF THE INVENTION

[0097] The present invention provides methods for treating cancer, particularly colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial cancer, using binding agents, including but not limited to polypeptides such as antibodies, that bind VEGF and/or DLL4 (e.g., a VEGF/DLL4-binding agent) optionally in combination with additional anti-cancer agent(s). The present invention also provides methods for treating cancer using therapeutic combinations, for example an anti-DLL4/VEGF bispecific antibody (e.g., 305B83) in combination with (a) leucovorin, 5-fluorouracil, and irinotecan, (b) paclitaxel, (c) gemcitabine and ABRAXANE®, or (c) paclitaxel and carboplatin. Compositions and kits including the VEGF/DLL4-binding agents and additional anti-cancer agent(s) are also provided. The methods of the invention include methods of inhibiting colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial tumor growth, methods of treating colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial cancer, methods of reducing tumorigenicity of colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial tumors, methods of reducing the frequency of cancer stem cells in colorectal, ovarian (e.g., platinum-resistant ovarian), pancreatic, and endometrial tumors, and/or methods of modulating angiogenesis in a patient with ovarian (e.g., platinum-resistant ovarian), colorectal, pancreatic, and endometrial cancer.

I. Definitions

[0098] To facilitate an understanding of the present invention, a number of terms and phrases are defined below.

[0099] The term "antibody" means an immunoglobulin molecule that recognizes and specifically binds a target, such as a protein, polypeptide, peptide, carbohydrate, polynucleotide, lipid, or combinations of the foregoing through at least one antigen recognition site or antigen-binding site within the variable region(s) of the immunoglobulin molecule. As used herein, the term "antibody" encompasses intact polyclonal antibodies, intact monoclonal antibodies, antibody fragments (such as Fab, Fab', F(ab')2, and Fv fragments), single chain Fv (scFv) mutants, multispecific antibodies such as bispecific antibodies, chimeric antibodies, humanized antibodies, human antibodies, fusion proteins comprising an antigen-binding site of an antibody, and any other modified immunoglobulin molecule comprising an antigen-binding site as long as the antibodies exhibit the desired biological activity. An antibody can be any of the five major classes of immunoglobulins: IgA, IgD, IgE, IgG, and IgM, or subclasses (isotypes) thereof (e.g., IgG1, IgG2, IgG3, IgG4, IgA1 and IgA2), based on the identity of their heavy chain constant domains referred to as alpha, delta, epsilon, gamma, and mu, respectively. The different classes of immunoglobulins have different and well known subunit structures and three-dimensional configurations. Antibodies can be naked or conjugated to other molecules including, but not limited to, toxins and radioisotopes.

[00100] The term "antibody fragment" refers to a portion of an intact antibody and as used herein refers to the antigenic determining variable regions or the antigen-binding site of an intact antibody. "Antibody fragment" as used herein comprises an antigen-binding site or epitope-binding site. Examples of antibody fragments include, but are not limited to Fab, Fab', $F(ab')_2$, and Fv fragments, linear antibodies, single chain antibodies, and multispecific antibodies formed from antibody fragments.

[00101] The term "variable region" of an antibody refers to the variable region of the antibody light chain or the variable region of the antibody heavy chain, either alone or in combination. The variable regions of the heavy chain and light chain generally consist of four framework regions connected by three complementarity determining regions (CDRs) (also known as hypervariable regions). The CDRs in each chain are held together in close proximity by the framework regions and, with the CDRs from the other chain, contribute to the formation of the antigen-binding site of the antibody. There are at least two techniques for determining CDRs: (1) an approach based on cross-species sequence variability (i.e., Kabat et al., 1991, *Sequences of Proteins of Immunological Interest, 5th Edition*, National Institutes of Health, Bethesda MD); and (2) an approach based on crystallographic studies of antigen-antibody complexes (Al-Lazikani et al., 1997, *J. Mol. Biol.* 273:927-948). In addition, combinations of these two approaches are sometimes used in the art to determine CDRs.

[00102] The term "monoclonal antibody" refers to a homogeneous antibody population involved in the highly specific recognition and binding of a single antigenic determinant or epitope. This is in contrast to polyclonal antibodies that typically include a mixture of different antibodies directed against a variety of different antigenic determinants. The term "monoclonal antibody" encompasses both intact and full-length monoclonal antibodies as well as antibody fragments (such as Fab, Fab', $F(ab')_2$, Fv fragments), single chain Fv (scFv) mutants, fusion proteins comprising an antibody portion, and any other modified immunoglobulin molecule comprising an antigen-binding site. Furthermore, "monoclonal antibody" refers to such antibodies made by any number of techniques, including but not limited to, hybridoma production, phage selection, recombinant expression, and transgenic animals.

[00103] The term "humanized antibody" refers to forms of non-human (e.g., murine) antibodies that are specific immunoglobulin chains, chimeric immunoglobulins, or fragments thereof that contain minimal non-human (e.g., murine) sequences.

[00104] The term "human antibody" means an antibody produced by a human or an antibody having an amino acid sequence corresponding to an antibody produced by a human made using any technique known in the art. This definition of a human antibody includes intact or full-length antibodies, and fragments thereof.

[00105] The term "chimeric antibodies" refers to antibodies wherein the amino acid sequence of the immunoglobulin molecule is derived from two or more species. Typically, the variable region of both light and heavy chains corresponds to the variable region of antibodies derived from one species of

mammal (e.g., mouse, rat, rabbit, etc.) with the desired specificity, affinity, and/or capability while the constant regions are homologous to the sequences in antibodies derived from another species (usually human) to avoid eliciting an immune response in that species.

[00106] The phrase "affinity-matured antibody" as used herein refers to an antibody with one or more alterations in one or more CDRs thereof that result in an improvement in the affinity of the antibody for antigen, compared to a parent antibody that does not possess those alterations(s). The definition also includes alterations in non-CDR residues made in conjunction with alterations to CDR residues. Desirable affinity-matured antibodies will have nanomolar or even picomolar affinities for the target antigen. Affinity-matured antibodies may be produced by techniques well-known in the art, including but not limited to, affinity maturation by heavy chain variable chain shuffling, light chain variable chain shuffling, random mutagenesis of CDR residues, random mutagenesis of framework residues, site-directed mutagenesis CDR residues, and site-directed mutagenesis of framework residues.

[00107] The terms "epitope" and "antigenic determinant" are used interchangeably herein and refer to that portion of an antigen capable of being recognized and specifically bound by a particular antibody. When the antigen is a polypeptide, epitopes can be formed both from contiguous amino acids and noncontiguous amino acids juxtaposed by tertiary folding of a protein. Epitopes formed from contiguous amino acids (also referred to as linear epitopes) are typically retained upon protein denaturing, whereas epitopes formed by tertiary folding (also referred to as conformational epitopes) are typically lost upon protein denaturing. An epitope typically includes at least 3, and more usually, at least 5 or 8-10 amino acids in a unique spatial conformation.

[00108] The terms "heteromultimeric molecule" or "heteromultimer" or "heteromultimeric complex" or "heteromultimeric polypeptide" are used interchangeably herein to refer to a molecule comprising at least a first polypeptide and a second polypeptide, wherein the second polypeptide differs in amino acid sequence from the first polypeptide by at least one amino acid residue. The heteromultimeric molecule can comprise a "heterodimer" formed by the first and second polypeptide or can form higher order tertiary structures where additional polypeptides are present.

[00109] The terms "antagonist" and "antagonistic" as used herein refer to any molecule that partially or fully blocks, inhibits, reduces, or neutralizes a biological activity of a target and/or signaling pathway (e.g., the Notch pathway). The term "antagonist" is used herein to include any molecule that partially or fully blocks, inhibits, reduces, or neutralizes the activity of a protein. Suitable antagonist molecules specifically include, but are not limited to, antagonist antibodies or antibody fragments.

[00110] The terms "modulation" and "modulate" as used herein refer to a change or an alteration in a biological activity. Modulation includes, but is not limited to, stimulating or inhibiting an activity. Modulation may be an increase or a decrease in activity (e.g., a decrease in angiogenesis or an increase in angiogenesis), a change in binding characteristics, or any other change in the biological, functional, or

immunological properties associated with the activity of a protein, pathway, or other biological point of interest.

[00111] The terms "selectively binds" or "specifically binds" mean that a binding agent or an antibody reacts or associates more frequently, more rapidly, with greater duration, with greater affinity, or with some combination of the above to the epitope, protein, or target molecule than with alternative substances, including unrelated proteins. In certain embodiments "specifically binds" means, for instance, that an antibody binds a protein with a K_D of about 0.1mM or less, but more usually less than about 1μM. In certain embodiments, "specifically binds" means that an antibody binds a target at times with a K_D of at least about 0.1μM or less, at other times at least about 0.01μM or less, and at other times at least about 1nM or less. Because of the sequence identity between homologous proteins in different species, specific binding can include an antibody that recognizes a protein in more than one species (e.g., human VEGF and mouse VEGF). Likewise, because of homology within certain regions of polypeptide sequences of different proteins, specific binding can include an antibody (or other polypeptide or binding agent) that recognizes more than one protein (e.g., human VEGF-A and human VEGF-B). It is understood that, in certain embodiments, an antibody or binding moiety that specifically binds a first target may or may not specifically bind a second target. As such, "specific binding" does not necessarily require (although it can include) exclusive binding, i.e. binding to a single target. Thus, an antibody may, in certain embodiments, specifically bind more than one target. In certain embodiments, multiple targets may be bound by the same antigen-binding site on the antibody. For example, an antibody may, in certain instances, comprise two identical antigen-binding sites, each of which specifically binds the same epitope on two or more proteins. In certain alternative embodiments, an antibody may be multispecific and comprise at least two antigen-binding sites with differing specificities. By way of non-limiting example, a bispecific antibody may comprise one antigen-binding site that recognizes an epitope on one protein (e.g., human VEGF) and further comprise a second, different antigen-binding site that recognizes a different epitope on a second protein (e.g., human DLL4). Generally, but not necessarily, reference to binding means specific binding.

[00112] The terms "polypeptide" and "peptide" and "protein" are used interchangeably herein and refer to polymers of amino acids of any length. The polymer may be linear or branched, it may comprise modified amino acids, and it may be interrupted by non-amino acids. The terms also encompass an amino acid polymer that has been modified naturally or by intervention; for example, disulfide bond formation, glycosylation, lipidation, acetylation, phosphorylation, or any other manipulation or modification, such as conjugation with a labeling component. Also included within the definition are, for example, polypeptides containing one or more analogs of an amino acid (including, for example, unnatural amino acids), as well as other modifications known in the art. It is understood that, because the polypeptides of this invention may be based upon antibodies, in certain embodiments, the polypeptides can occur as single chains or associated chains.

[00113] The terms "polynucleotide" and "nucleic acid" are used interchangeably herein and refer to polymers of nucleotides of any length, and include DNA and RNA. The nucleotides can be deoxyribonucleotides, ribonucleotides, modified nucleotides or bases, and/or their analogs, or any substrate that can be incorporated into a polymer by DNA or RNA polymerase.

[00114] "Conditions of high stringency" may be identified by those that: (1) employ low ionic strength and high temperature for washing, for example 15mM sodium chloride/1.5mM sodium citrate/0.1% sodium dodecyl sulfate at 50°C; (2) employ during hybridization a denaturing agent, such as formamide, for example, 50% (v/v) formamide with 0.1% bovine serum albumin/0.1% Ficoll/0.1% polyvinylpyrrolidone/50mM sodium phosphate buffer at pH 6.5 in 5x SSC (0.75M NaCl, 75mM sodium citrate) at 42°C; or (3) employ during hybridization 50% formamide in 5x SSC, 50mM sodium phosphate (pH 6.8), 0.1% sodium pyrophosphate, 5x Denhardt's solution, sonicated salmon sperm DNA (50µg/ml), 0.1% SDS, and 10% dextran sulfate at 42°C, with washes at 42°C in 0.2x SSC and 50% formamide, followed by a high-stringency wash consisting of 0.1x SSC containing EDTA at 55°C.

[00115] The terms "identical" or percent "identity" in the context of two or more nucleic acids or polypeptides, refer to two or more sequences or subsequences that are the same or have a specified percentage of nucleotides or amino acid residues that are the same, when compared and aligned (introducing gaps, if necessary) for maximum correspondence, not considering any conservative amino acid substitutions as part of the sequence identity. The percent identity may be measured using sequence comparison software or algorithms or by visual inspection. Various algorithms and software that may be used to obtain alignments of amino acid or nucleotide sequences are well-known in the art. These include, but are not limited to, BLAST, ALIGN, Megalign, BestFit, GCG Wisconsin Package, and variations thereof. In some embodiments, two nucleic acids or polypeptides of the invention are substantially identical, meaning they have at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, and in some embodiments at least 95%, 96%, 97%, 98%, 99% nucleotide or amino acid residue identity, when compared and aligned for maximum correspondence, as measured using a sequence comparison algorithm or by visual inspection. In some embodiments, identity exists over a region of the sequences that is at least about 10, at least about 20, at least about 40-60 residues, at least about 60-80 residues in length or any integral value therebetween. In some embodiments, identity exists over a longer region than 60-80 residues, such as at least about 80-100 residues, and in some embodiments the sequences are substantially identical over the full length of the sequences being compared, such as the coding region of a nucleotide sequence.

[00116] A "conservative amino acid substitution" is one in which one amino acid residue is replaced with another amino acid residue having a similar side chain. Families of amino acid residues having similar side chains have been defined in the art, including basic side chains (e.g., lysine, arginine, histidine), acidic side chains (e.g., aspartic acid, glutamic acid), uncharged polar side chains (e.g., glycine,

asparagine, glutamine, serine, threonine, tyrosine, cysteine), nonpolar side chains (e.g., alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tryptophan), beta-branched side chains (e.g., threonine, valine, isoleucine) and aromatic side chains (e.g., tyrosine, phenylalanine, tryptophan, histidine). For example, substitution of a phenylalanine for a tyrosine is a conservative substitution. Preferably, conservative substitutions in the sequences of the polypeptides and antibodies of the invention do not abrogate the binding of the polypeptide or antibody containing the amino acid sequence, to the antigen to which the polypeptide or antibody binds. Methods of identifying nucleotide and amino acid conservative substitutions which do not eliminate antigen binding are well-known in the art.

[00117] The term "vector" as used herein means a construct, which is capable of delivering, and usually expressing, one or more gene(s) or sequence(s) of interest in a host cell. Examples of vectors include, but are not limited to, viral vectors, naked DNA or RNA expression vectors, plasmid, cosmid, or phage vectors, DNA or RNA expression vectors associated with cationic condensing agents, and DNA or RNA expression vectors encapsulated in liposomes.

[00118] A polypeptide, antibody, polynucleotide, vector, cell, or composition which is "isolated" is a polypeptide, antibody, polynucleotide, vector, cell, or composition which is in a form not found in nature. Isolated polypeptides, antibodies, polynucleotides, vectors, cells, or compositions include those which have been purified to a degree that they are no longer in a form in which they are found in nature. In some embodiments, a polypeptide, antibody, polynucleotide, vector, cell, or composition which is isolated is substantially pure.

[00119] The term "substantially pure" as used herein refers to material which is at least 50% pure (i.e., free from contaminants), at least 90% pure, at least 95% pure, at least 98% pure, or at least 99% pure.

[00120] The terms "cancer" and "cancerous" as used herein refer to or describe the physiological condition in mammals in which a population of cells are characterized by unregulated cell growth.

[00121] The terms "tumor" and "neoplasm" as used herein refer to any mass of tissue that results from excessive cell growth or proliferation, either benign (noncancerous) or malignant (cancerous) including pre-cancerous lesions.

[00122] The term "metastasis" as used herein refers to the process by which a cancer spreads or transfers from the site of origin to other regions of the body with the development of a similar cancerous lesion at a new location. A "metastatic" or "metastasizing" cell is one that loses adhesive contacts with neighboring cells and migrates via the bloodstream or lymph from the primary site of disease to invade neighboring body structures.

[00123] The terms "cancer stem cell" and "CSC" and "tumor stem cell" and "tumor initiating cell" are used interchangeably herein and refer to cells from a cancer or tumor that: (1) have extensive proliferative capacity; 2) are capable of asymmetric cell division to generate one or more types of differentiated cell progeny wherein the differentiated cells have reduced proliferative or developmental potential; and (3) are

capable of symmetric cell divisions for self-renewal or self-maintenance. These properties confer on the cancer stem cells the ability to form or establish a tumor or cancer upon serial transplantation into an immunocompromised host (e.g., a mouse) compared to the majority of tumor cells that fail to form tumors. Cancer stem cells undergo self-renewal versus differentiation in a chaotic manner to form tumors with abnormal cell types that can change over time as mutations occur.

[00124] The terms "cancer cell" and "tumor cell" refer to the total population of cells derived from a cancer or tumor or pre-cancerous lesion, including both non-tumorigenic cells, which comprise the bulk of the cancer cell population, and tumorigenic stem cells (cancer stem cells). As used herein, the terms "cancer cell" or "tumor cell" will be modified by the term "non-tumorigenic" when referring solely to those cells lacking the capacity to renew and differentiate to distinguish those tumor cells from cancer stem cells.

[00125] The term "tumorigenic" as used herein refers to the functional features of a cancer stem cell including the properties of self-renewal (giving rise to additional tumorigenic cancer stem cells) and proliferation to generate all other tumor cells (giving rise to differentiated and thus non-tumorigenic tumor cells).

[00126] The term "tumorigenicity" as used herein refers to the ability of a random sample of cells from the tumor to form palpable tumors upon serial transplantation into immunocompromised hosts (e.g., mice). This definition also includes enriched and/or isolated populations of cancer stem cells that form palpable tumors upon serial transplantation into immunocompromised hosts (e.g., mice).

[00127] The term "platinum-resistant" in the context of ovarian cancer, refers to a patient with recurrent disease having no response to platinum-based chemotherapy (i.e., disease progression or stable disease as the best response) or, if the cancer did initially respond to platinum-based chemotherapy, but recurred within 6 months of primary treatment. Most patients with recurrent ovarian cancer eventually develop platinum resistance.

[00128] The term "subject" refers to any animal (e.g., a mammal), including, but not limited to, humans, non-human primates, canines, felines, rodents, and the like, which is to be the recipient of a particular treatment. Typically, the terms "subject" and "patient" are used interchangeably herein in reference to a human subject.

[00129] The term "pharmaceutically acceptable" refers to a product or compound approved (or approvable) by a regulatory agency of the Federal government or a state government or listed in the U.S. Pharmacopeia or other generally recognized pharmacopeia for use in animals, including humans.

[00130] The terms "pharmaceutically acceptable excipient, carrier or adjuvant" or "acceptable pharmaceutical carrier" refer to an excipient, carrier or adjuvant that can be administered to a subject, together with at least one binding agent (e.g., an antibody) of the present disclosure, and which does not

destroy the activity of the binding agent. The excipient, carrier or adjuvant should be nontoxic when administered with a binding agent in doses sufficient to deliver a therapeutic effect.

[00131] The terms "effective amount" or "therapeutically effective amount" or "therapeutic effect" refer to an amount of a binding agent, an antibody, polypeptide, polynucleotide, small organic molecule, or other drug effective to "treat" a disease or disorder in a subject or mammal. In the case of cancer, the therapeutically effective amount of a drug (e.g., an antibody) has a therapeutic effect and as such can reduce the number of cancer cells; decrease tumorigenicity, tumorigenic frequency or tumorigenic capacity; reduce the number or frequency of cancer stem cells; reduce the tumor size; reduce the cancer cell population; inhibit and/or stop cancer cell infiltration into peripheral organs including, for example, the spread of cancer into soft tissue and bone; inhibit and/or stop tumor or cancer cell metastasis; inhibit and/or stop tumor or cancer cell growth; relieve to some extent one or more of the symptoms associated with the cancer; reduce morbidity and mortality; improve quality of life; or a combination of such effects. To the extent the agent, for example an antibody, prevents growth and/or kills existing cancer cells, it can be referred to as cytostatic and/or cytotoxic.

[00132] The terms "treating" or "treatment" or "to treat" or "alleviating" or "to alleviate" refer to both 1) therapeutic measures that cure, slow down, lessen symptoms of, and/or halt progression of a diagnosed pathologic condition or disorder and 2) prophylactic or preventative measures that prevent or slow the development of a targeted pathologic condition or disorder. Thus those in need of treatment include those already with the disorder; those prone to have the disorder; and those in whom the disorder is to be prevented. In some embodiments, a subject is successfully "treated" according to the methods of the present invention if the patient shows one or more of the following: a reduction in the number of or complete absence of cancer cells; a reduction in the tumor size; inhibition of or an absence of cancer cell infiltration into peripheral organs including the spread of cancer cells into soft tissue and bone; inhibition of or an absence of tumor or cancer cell metastasis; inhibition or an absence of cancer growth; relief of one or more symptoms associated with the specific cancer; reduced morbidity and mortality; improvement in quality of life; reduction in tumorigenicity; reduction in the number or frequency of cancer stem cells; or some combination of effects.

[00133] By "FOLFIRI" is meant the combination of leucovorin (LV), 5-fluorouracil (FU), and irinotecan where the *l*-LV 200 mg/m^2 or *dl*-LV 400 mg/m^2 is given as a 2-hour infusion, and the irinotecan at 180 mg/m^2 is given as a 90-minute infusion in 500 mL dextrose 5% at the same time (e.g., by a Y connector), followed by bolus FU 400 mg/m^2 and a 46-hour infusion FU at 2,400 mg/m^2-3,000 mg/m^2 given every 2 weeks.

[00134] By "pancreatic cancer" or "pancreatic tumor" is meant any cancer or tumor that originally develops in the pancreas. The most common type of pancreatic cancer is pancreatic adenocarcinoma. Other types of pancreatic cancer include islet cell carcinoma, pancreaticoblastoma, and ampullary cancer.

[00135] By "colorectal cancer" or "colorectal tumor" is meant any cancer that develops in large intestine, i.e., the colon or rectum. The most colorectal cancers are adenocarcinomas. Other types of colorectal cancer include carcinoid tumors, gastrointestinal stromal tumors, and sarcomas.

[00136] By "ovarian cancer" is meant any cancer that develops in the ovaries, fallopian tubes, or primary peritoneum and spreads to the ovaries. The most common ovarian cancer is ovarian epithelial cancer. Other ovarian cancers include germ cell cancers.

[00137] By "endometrial cancer" is meant any cancer that develops in the uterine lining. Endometrial cancers include endometrial carcinomas, for example, adenocarcinomas, carcinosarcomas, squamous cell carcinomas, undifferentiated carcinomas, small cell carcinomas, and transitional carcinomas, the most common of which are adenocarcinomas.

[00138] As used in the present disclosure and claims, the singular forms "a", "an", and "the" include plural forms unless the context clearly dictates otherwise.

[00139] It is understood that wherever embodiments are described herein with the language "comprising" otherwise analogous embodiments described in terms of "consisting of" and/or "consisting essentially of" are also provided. It is also understood that wherever embodiments are described herein with the language "consisting essentially of" otherwise analogous embodiments described in terms of "consisting of" are also provided.

[00140] The term "and/or" as used in a phrase such as "A and/or B" herein is intended to include both A and B; A or B; A (alone); and B (alone). Likewise, the term "and/or" as used in a phrase such as "A, B, and/or C" is intended to encompass each of the following embodiments: A, B, and C; A, B, or C; A or C; A or B; B or C; A and C; A and B; B and C; A (alone); B (alone); and C (alone).

II. Methods of use and pharmaceutical compositions

[00141] The binding agents (including polypeptides and antibodies) of the invention that bind (e.g., specifically bind) VEGF and/or DLL4 are useful in a variety of applications including, but not limited to, therapeutic treatment methods, such as the treatment of cancer, particularly in combination with leucovorin, 5-fluorouracil, and irinotecan (e.g., for treatment of colorectal cancer), in combination with paclitaxel (e.g., for treatment of ovarian cancer such as platinum-resistant ovarian cancer), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of pancreatic cancer), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of endometrial cancer). In certain embodiments, the tumor is platinum-resistant ovarian cancer. In some embodiments, the cancer is endometrial cancer. In certain embodiments, the agents are useful for inhibiting VEGF activity, inhibiting DLL4-induced Notch signaling, inhibiting tumor growth, reducing tumor volume, reducing the frequency of cancer stem cells in the tumor, reducing the tumorigenicity of the tumor, modulating angiogenesis in a patient with the tumor, and/or inhibiting angiogenesis in a patient with a tumor. The methods of use may be *in vitro*, *ex vivo*, or

in vivo. In certain embodiments, a VEGF/DLL4-binding agent is an antagonist of human VEGF. In certain embodiments, a VEGF/DLL4-binding agent is an antagonist of human DLL4. In certain embodiments, a VEGF/DLL4-binding agent is an antagonist of both human VEGF and human DLL4.

[00142] The present invention provides methods for inhibiting growth of a tumor using the VEGF/DLL4-binding agents or antibodies described herein, particularly in combination with FOLFIRI (e.g., for treatment of a colorectal tumor) in combination with paclitaxel (e.g., for treatment of an ovarian tumor such as a platinum-resistant ovarian tumor), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of a pancreatic tumor), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of an endometrial tumor). In certain embodiments, the tumor is a platinum-resistant ovarian tumor. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the method of inhibiting growth of a tumor comprises contacting a tumor cell with a VEGF/DLL4-binding agent (e.g., antibody) *in vitro*. For example, an immortalized cell line or a cancer cell line is cultured in medium to which is added an anti-VEGF antibody, an anti-DLL4 antibody, an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination to inhibit tumor cell growth. In some embodiments, tumor cells are isolated from a patient sample such as, for example, a tissue biopsy or blood sample and cultured in medium to which is added a VEGF/DLL4-binding agent or therapeutic combination to inhibit tumor cell growth.

[00143] In some embodiments, the method of inhibiting growth of a tumor comprises contacting a tumor or tumor cell with a VEGF/DLL4-binding agent (e.g., antibody) *in vivo*, particularly in combination with FOLFIRI (e.g., for treatment of a colorectal tumor), in combination with paclitaxel (e.g., for treatment of an ovarian tumor such as platinum-resistant ovarian cancer), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of a pancreatic tumor), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of an endometrial tumor). In certain embodiments, the tumor is a platinum-resistant ovarian tumor. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, contacting a tumor or tumor cell with a VEGF/DLL4-binding agent is undertaken in an animal model. For example, an anti-VEGF antibody, an anti-DLL4 antibody, an anti-VEGF/anti-DLL4 bispecific antibody, or therapeutic combination may be administered to an immunocompromised host animal (e.g., NOD/SCID mice) which has a tumor xenograft. In some embodiments, tumor cells and/or cancer stem cells are isolated from a patient sample such as, for example, a tissue biopsy or blood sample and injected into an immunocompromised host animal (e.g., NOD/SCID mice) that is then administered a VEGF/DLL4-binding agent or therapeutic combination to inhibit tumor cell growth. In some embodiments, the VEGF/DLL4-binding agent or therapeutic combination is administered at the same time or shortly after introduction of tumorigenic cells into the animal to prevent tumor growth ("preventative model"). In some embodiments, the VEGF/DLL4-binding agent or therapeutic combination is administered as a therapeutic after tumors have grown to a specified size ("therapeutic model"). In certain

embodiments, the VEGF/DLL4-binding agent is a bispecific antibody that specifically binds human VEGF and human DLL4.

[00144] In certain embodiments, the method of inhibiting growth of a tumor comprises administering to a subject a therapeutically effective amount of a VEGF/DLL4-binding agent, particularly in combination with FOLFIRI (e.g., for treatment of a colorectal tumor), in combination with paclitaxel (e.g., for treatment of ovarian cancer such as platinum-resistant ovarian cancer), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of pancreatic cancer), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of an endometrial tumor). In certain embodiments, the tumor is a platinum-resistant ovarian tumor. In certain embodiments, the tumor is an endometrial tumor. In certain embodiments, the subject is a human. In certain embodiments, the subject has a tumor (e.g., pancreatic, colorectal or endometrial tumor) or has had at least a portion of a tumor surgically removed. In certain embodiments, the tumor comprises cancer stem cells. In certain embodiments, the frequency of cancer stem cells in the tumor is reduced by administration of the VEGF/DLL4-binding agent or therapeutic combination. The invention also provides a method of reducing the frequency of cancer stem cells in a tumor (e.g., colorectal, ovarian, pancreatic, or endometrial tumor), comprising contacting the tumor with an effective amount of a VEGF/DLL4-binding agent (e.g., an anti-VEGF/anti-DLL4 bispecific antibody) or therapeutic combination. In some embodiments, a method of reducing the frequency of cancer stem cells in a tumor, comprises administering to a subject who has a tumor a therapeutically effective amount of a VEGF/DLL4-binding agent or therapeutic combination.

[00145] The present invention further provides methods for treating cancer comprising administering a therapeutically effective amount of a VEGF/DLL4-binding agent to a subject, particularly in combination with FOLFIRI (e.g., for treatment of a colorectal tumor), in combination with paclitaxel (e.g., for treatment of ovarian cancer such as platinum-resistant ovarian cancer), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of pancreatic cancer), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of an endometrial tumor). In certain embodiments, the tumor is a platinum-resistant ovarian tumor. In certain embodiments, the tumor is an endometrial tumor. In some embodiments, the VEGF/DLL4-binding agent binds VEGF, and inhibits or reduces growth of the cancer (e.g., colorectal, ovarian, pancreatic, or endometrial cancer). In some embodiments, the VEGF/DLL4-binding agent binds DLL4, and inhibits or reduces growth of the cancer. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody that binds VEGF and DLL4, and inhibits or reduces growth of the cancer. In some embodiments, the VEGF/DLL4-binding agent binds VEGF, interferes with VEGF/VEGF receptor interactions, and inhibits or reduces growth of the cancer. In some embodiments, the VEGF/DLL4-binding agent binds DLL4, interferes with DLL4/Notch interactions, and inhibits or reduces growth of the cancer. In some embodiments, the VEGF/DLL4-binding agent binds both VEGF and DLL4, interferes with VEGF/VEGF receptor interactions and with DLL4/Notch interactions, and

inhibits or reduces growth of the cancer. In some embodiments, the VEGF/DLL4-binding agent binds DLL4, and reduces the frequency of cancer stem cells in the cancer.

[00146] The present invention provides methods of treating cancer comprising administering a therapeutically effective amount of a VEGF/DLL4-binding agent to a subject (e.g., a subject in need of treatment), particularly in combination with FOLFIRI (e.g., for treatment of a colorectal tumor), in combination with paclitaxel (e.g., for treatment of ovarian cancer such as platinum-resistant ovarian cancer), in combination with gemcitabine and nab-paclitaxel (e.g., for treatment of pancreatic cancer), and in combination with paclitaxel and/or carboplatin (e.g., for treatment of an endometrial tumor). In certain embodiments, the tumor is a platinum-resistant ovarian tumor. In certain embodiments, the cancer is endometrial cancer. In certain embodiments, the subject is a human. In certain embodiments, the subject has a cancerous tumor. In certain embodiments, the subject has had at least a portion of a tumor (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor) surgically removed.

[00147] The subject's cancer/tumor, may, in some embodiments, be refractory to certain treatment(s). In some embodiments, the subject's cancer (or tumor) may be chemorefractory. In those cases, the therapy provided herein can be second-line or third-line therapy for the cancer/tumor. In certain embodiments, the subject's cancer may be resistant to anti-VEGF therapy or anti-DLL4 therapy.

[00148] In certain embodiments of any of the methods described herein, the VEGF/DLL4-binding agent is a bispecific antibody that specifically binds human VEGF and human DLL4. In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising

YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00149] In certain embodiments of any of the methods described herein, the VEGF/DLL4 bispecific antibody comprises a first heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64, and a first and a second light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In some embodiments, the VEGF/DLL4 bispecific antibody comprises a first heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:64, and a first and a second light chain variable region having at least 80% sequence identity to SEQ ID NO:12.

[00150] In some embodiments of any of the methods described herein, the VEGF/DLL4-binding agent is an antibody. In some embodiments, the VEGF/DLL4-binding agent is an anti-VEGF antibody. In some embodiments, the anti-VEGF antibody is antibody 219R45. In some embodiments, the VEGF/DLL4-binding agent is an anti-DLL4 antibody. In some embodiments, the anti-DLL4 antibody is antibody 21R79, antibody 21R83, or antibody 219R45. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising an antigen-binding site from antibody 21R79, antibody 21R75, or antibody 21R83. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising a first antigen-binding site from antibody 219R45 and a second antigen-binding site from antibody 21R79, antibody 21M18, antibody 21R75, or antibody 21R83. In some embodiments, the VEGF/DLL4-binding agent is the bispecific antibody 219R45-MB-21M18 (305B18). In some embodiments, the VEGF/DLL4-binding agent is the bispecific antibody 219R45-MB-21R79 (305B79). In some embodiments, the VEGF/DLL4-binding agent is the bispecific antibody 219R45-MB-21R75 (305B75). In some embodiments, the VEGF/DLL4-binding agent is the bispecific antibody 219R45-MB-21R83 (305B83).

[00151] The present invention further provides pharmaceutical compositions comprising the binding agents in combination with an additional therapeutic agent (e.g., those described herein). In certain embodiments, the pharmaceutical compositions further comprise a pharmaceutically acceptable vehicle. These pharmaceutical compositions find use in inhibiting tumor growth (e.g., colorectal, ovarian, pancreatic, or endometrial tumor growth) and/or treating cancer (e.g., colorectal, ovarian, pancreatic, or endometrial cancer) in a subject (e.g., a human patient).

[00152] In certain embodiments, the invention provides pharmaceutical compositions comprising bispecific antibodies, wherein at least about 90%, at least about 95%, at least about 98%, at least about 99% of the antibodies in the composition are bispecific antibodies or heterodimeric antibodies. In certain

embodiments, the bispecific antibodies are IgG (e.g., IgG2 or IgG1) antibodies. In certain embodiments, less than about 10%, less than about 5%, less than about 2% or less than about 1% of the total antibodies in the compositions are monospecific antibodies or homodimeric antibodies. In certain embodiments, the antibodies in the composition are at least about 98% heterodimeric.

[00153] In certain embodiments, formulations are prepared for storage and use by combining a purified antibody, agent, or therapeutic combination of the present invention with a pharmaceutically acceptable vehicle (e.g., a carrier or excipient). Suitable pharmaceutically acceptable vehicles include, but are not limited to, non-toxic buffers such as phosphate, citrate, and other organic acids; salts such as sodium chloride; antioxidants including ascorbic acid and methionine; preservatives such as octadecyldimethylbenzyl ammonium chloride, hexamethonium chloride, benzalkonium chloride, benzethonium chloride, phenol, butyl or benzyl alcohol, alkyl parabens, such as methyl or propyl paraben, catechol, resorcinol, cyclohexanol, 3-pentanol, and m-cresol; low molecular weight polypeptides (e.g., less than about 10 amino acid residues); proteins such as serum albumin, gelatin, or immunoglobulins; hydrophilic polymers such as polyvinylpyrrolidone; amino acids such as glycine, glutamine, asparagine, histidine, arginine, or lysine; carbohydrates such as monosaccharides, disaccharides, glucose, mannose, or dextrins; chelating agents such as EDTA; sugars such as sucrose, mannitol, trehalose or sorbitol; salt-forming counter-ions such as sodium; metal complexes such as Zn-protein complexes; and non-ionic surfactants such as TWEEN or polyethylene glycol (PEG). (*Remington: The Science and Practice of Pharmacy, 22st Edition,* 2012, Pharmaceutical Press, London).

[00154] The pharmaceutical compositions of the present invention can be administered in any number of ways for either local or systemic treatment. Administration can be topical by epidermal or transdermal patches, ointments, lotions, creams, gels, drops, suppositories, sprays, liquids, and powders; pulmonary by inhalation or insufflation of powders or aerosols, including by nebulizer, intratracheal, and intranasal; oral; or parenteral including intravenous, intraarterial, intratumoral, subcutaneous, intraperitoneal, intramuscular (e.g., injection or infusion), or intracranial (e.g., intrathecal or intraventricular).

[00155] The therapeutic formulation can be in unit dosage form. Such formulations include tablets, pills, capsules, powders, granules, solutions or suspensions in water or non-aqueous media, or suppositories. In solid compositions such as tablets the principal active ingredient is mixed with a pharmaceutical carrier. Conventional tableting ingredients include corn starch, lactose, sucrose, sorbitol, talc, stearic acid, magnesium stearate, dicalcium phosphate or gums, and diluents (e.g., water). These can be used to form a solid preformulation composition containing a homogeneous mixture of a compound of the present invention, or a non-toxic pharmaceutically acceptable salt thereof. The solid preformulation composition is then subdivided into unit dosage forms of a type described above. The tablets, pills, etc. of the formulation or composition can be coated or otherwise compounded to provide a dosage form affording the advantage of prolonged action. For example, the tablet or pill can comprise an inner composition

covered by an outer component. Furthermore, the two components can be separated by an enteric layer that serves to resist disintegration and permits the inner component to pass intact through the stomach or to be delayed in release. A variety of materials can be used for such enteric layers or coatings, such materials include a number of polymeric acids and mixtures of polymeric acids with such materials as shellac, cetyl alcohol and cellulose acetate.

[00156] The VEGF/DLL4-binding agents, antibodies, and therapeutic combinations described herein can also be entrapped in microcapsules. Such microcapsules are prepared, for example, by coacervation techniques or by interfacial polymerization, for example, hydroxymethylcellulose or gelatin-microcapsules and poly-(methylmethacylate) microcapsules, respectively, in colloidal drug delivery systems (for example, liposomes, albumin microspheres, microemulsions, nanoparticles and nanocapsules) or in macroemulsions as described in *Remington: The Science and Practice of Pharmacy, 22st Edition,* 2012, Pharmaceutical Press, London.

[00157] In certain embodiments, pharmaceutical formulations include a VEGF/DLL4-binding agent (e.g., an antibody) or therapeutic combination of the present invention complexed with liposomes. Methods to produce liposomes are known to those of skill in the art. For example, some liposomes can be generated by reverse phase evaporation with a lipid composition comprising phosphatidylcholine, cholesterol, and PEG-derivatized phosphatidylethanolamine (PEG-PE). Liposomes can be extruded through filters of defined pore size to yield liposomes with the desired diameter.

[00158] In certain embodiments, sustained-release preparations can be produced. Suitable examples of sustained-release preparations include semi-permeable matrices of solid hydrophobic polymers containing a VEGF/DLL4-binding agent (e.g., an antibody) or therapeutic combination, where the matrices are in the form of shaped articles (e.g., films or microcapsules). Additional examples of sustained-release matrices include polyesters, hydrogels such as poly(2-hydroxyethyl-methacrylate) or poly(vinyl alcohol), polylactides, copolymers of L-glutamic acid and 7 ethyl-L-glutamate, non-degradable ethylene-vinyl acetate, degradable lactic acid-glycolic acid copolymers such as the LUPRON DEPOT™ (injectable microspheres composed of lactic acid-glycolic acid copolymer and leuprolide acetate), sucrose acetate isobutyrate, and poly-D-(-)-3-hydroxybutyric acid.

III. Combination with gemcitabine and ABRAXANE®

[00159] In certain embodiments, the VEGF/DLL4 binding agent (e.g., anti VEGF/DLL4 bispecific antibody such as 305B83) is administered in combination with gemcitabine and ABRAXANE®. The combination that includes gemcitabine and ABRAXANE® can be used, for example, to treat pancreatic cancer. The precise dosing and timing can be determined by a physician or can be any of the dosing regimens described herein. In particular embodiments, the VEGF/DLL4 binding agent is administered

within three months, two months, one month, three weeks, two weeks, one week, three days, two days, or one day of the gemictiabine and ABRAXANE®.

[00160] The VEGF/DLL4 binding agent (e.g., 305B83) may be administered at a dose between 0.1 mg/kg and 20 mg/kg or between 0.5 mg/kg and 10 mg/kg or about 0.5, 1, 2.5, 4, 5, 10, or 15 mg/kg. In some embodiments, the dose is 3 mg/kg, 5 mg/kg, 10 mg/kg, or 15 mg/kg. In some embodiments, the dose is about 1 mg/kg, 2.5 mg/kg, or 5 mg/kg. In some embodiments, the dose is administered about every two weeks or about every three weeks. In other embodiments, the dose is administered about every week, every ten days, every four weeks, every six weeks, or every two months.

[00161] In certain embodiments, the ABRAXANE® is provided at a dose between 50 and 300 mg/m^2, for example, at about 50, 100, 125, 150, 175, 200, 225, 250, 260, 275, or 300 mg/m^2 administered every week, every other week, every three weeks, on days, 1, 8, and 15 of a 21-day cycle, or on days 1, 8, and 15 of a 28-day cycle. Typically, ABRAXANE® is delivered as an intravenous infusion, e.g., over 20-60 minutes, e.g., 30-40 min or about 30 min. In certain embodiments, ABRAXANE® is administered at a dose of about 260 mg/m^2 intravenously over about 30 minutes every 3 weeks or at a dose of about 100 mg/m^2 intravenously over about 30 minutes on days 1, 8, and 15 of each 21-day cycle.

[00162] In a particular embodiment, ABRAXANE® is administered at a about 125 mg/m^2 intravenously over 30-40 minutes on days 1, 8, and 15 of each 28-day cycle, and gemcitabine is administered on days 1, 8 and 15 of each 28-day cycle immediately after ABRAXANE®. In these embodiments, gemcitabine can be dosed at about 1000 mg/m^2 intravenously for about 30 minutes.

[00163] In some embodiments, gemcitabine is dosed at 100-2000 mg/m^2, for example, at about 100, 200, 300, 500, 700, 1000, 1250, 1500, or 2000 mg/m^2 intravenously, e.g., infused from 20-60 minutes or about 30 minutes.

IV. Combination with leucovorin, fluorouracil, and irinotecan

[00164] In certain embodiments, the VEGF/DLL4 binding agent (e.g., anti VEGF/DLL4 bispecific antibody such as 305B83) is administered in combination with leucovorin, fluorouracil, and irinotecan. This combination can be used to treat colorectal cancer (e.g., metastatic colorectal cancer). The precise dosing and timing can be determined by a physician or can be any of the dosing regimens described herein. In particular embodiments, the VEGF/DLL4 binding agent is administered within three months, two months, one month, three weeks, two weeks, one week, three days, two days, or one day of the leucovorin, fluorouracil, and irinotecan.

[00165] The VEGF/DLL4 binding agent (e.g., 305B83) may be administered at a dose between 0.1 mg/kg and 20 mg/kg or between 0.5 mg/kg and 10 mg/kg or about 0.5, 1, 2.5, 3, 4, 5, 10, or 15 mg/kg. In some embodiments, the dose is about 3 mg/kg, 5 mg/kg, 10 mg/kg, or 15 mg/kg. In some embodiments, the dose is about 1 mg/kg, 2.5 mg/kg, or 5 mg/kg. In some embodiments, the dose is administered every

about every two weeks or about every three weeks. In other embodiments, the dose is administered about every week, every ten days, every four weeks, every six weeks, or every two months.

[00166] In certain embodiments, the timing and dosing follows that described in Tournigand et al., *J. Clin. Oncol.* 22:229-237, 2004. In these embodiments, the agents are given as the "FOLFIRI" combination, i.e., where the *l*-leucovorin 200 mg/m^2 or *dl*-leucovorin 400 mg/m^2 is given as a 2-hour infusion, and the irinotecan at 180 mg/m^2 is given as a 90-minute infusion in 500 ml dextrose 5% at the same time (e.g., by a Y connector), followed by bolus fluorouracil 400 mg/m^2 and a 46-hour infusion fluorouracil at 2,400 mg/m^2-3,000 mg/m^2 given every 2 weeks.

[00167] In other embodiments, the *dl*-leucovorin can be given at 100-700 mg/m^2, e.g., at about 100, 200, 250, 300, 350, 400, 450, 500, 600, or 700 mg/m^2 (or at the appropriate equivalent dosing if *l*-leucovorin is used). The leucovorin can be administered by intravenous infusion, for example, over 0.5-3 hours, e.g., about 0.5, 1, 1.5, 2, 2.5, or 3 hours. The irinotecan can be administered at a dose of 50-300 mg/m^2, e.g., at about 100, 125, 150, 175, 180, 200, 225, 250, 275, or 300 mg/m^2. The irinotecan can be administered over 0.5-3 hours, e.g., about 0.5, 1, 1.5, 2, 2.5, or 3 hours. The total fluorouracil dosage can be 1000-5000 mg/m^2, e.g., about 1000, 1250, 1500, 2000, 2500, 3000, 3500, 4000 4500, or 5000 mg/m^2. The drugs can be administered, for example, twice weekly, weekly, once every other week, once every three weeks, or once every four weeks.

[00168] In certain embodiments, oxaliplatin can be administered in place of or in addition to the irinotecan. For example, the oxaliplatin may be administered as part of the FOLFOX4, FOLFOX6, or FOLFIRINOX dosing schedule.

V. Combination with paclitaxel

[00169] In certain embodiments, the VEGF/DLL4 binding agent (e.g., anti VEGF/DLL4 bispecific antibody such as 305B83) is administered in combination with paclitaxel. This combination can be used to treat ovarian cancer (e.g., platinum-resistant ovarian cancer). In certain embodiments, the cancer has been treated with two or more prior therapies (e.g., three prior therapies or four prior therapies) and/or has been previously treated with an anti-VEGF agent, such as bevacizumab. The precise dosing and timing can be determined by a physician or can be any of the dosing regimens described herein. In particular embodiments, the VEGF/DLL4 binding agent is administered within three months, two months, one month, three weeks, two weeks, one week, three days, two days, or one day of the paclitaxel. The VEGF/DLL4 binding agent (e.g., 305B83) may be administered at a dose between 0.1 mg/kg and 20 mg/kg or between 0.5 mg/kg and 10 mg/kg, or about 0.5, 1, 2.5, 3, 4, 5, 10, or 15 mg/kg. In some embodiments, the dose is 3 mg/kg, 5 mg/kg, 10 mg/kg, or 15 mg/kg. In some embodiments, the dose is 1 mg/kg, 2.5 mg/kg, or 5 mg/kg. In some embodiments, the dose is administered every about every two

weeks or about every three weeks. In other embodiments, the dose is administered every week, every ten days, every four weeks, every six weeks or every two months.

[00170] Paclitaxel can be given at a dose of 175 mg/m^2, but also be in the range of 50-300 mg/m^2, for example, about 50, 75, 100, 125, 135, 150, 175, 200, 225, 250, or 300 mg/m^2. The paclitaxel can be dosed every week, every two weeks, every three weeks, every four weeks, every month, every six weeks, or every two months.

V. Combination with paclitaxel and carboplatin

[00171] In certain embodiments, the VEGF/DLL4 binding agent (e.g., anti VEGF/DLL4 bispecific antibody such as 305B83) is administered in combination with paclitaxel and carboplatin. This combination can be used to treat endometrial cancer. The precise dosing and timing can be determined by a physician or can be any of the dosing regimens described herein. In particular embodiments, the VEGF/DLL4 binding agent is administered within three months, two months, one month, three weeks, two weeks, one week, three days, two days, or one day of the paclitaxel and carboplatin.

[00172] The VEGF/DLL4 binding agent (e.g., 305B83) may be administered at a dose between 0.1 mg/kg and 20 mg/kg or between 0.5 mg/kg and 10 mg/kg or about 0.5, 1, 2.5, 3, 4, 5, 10, or 15 mg/kg. In some embodiments, the dose is 3 mg/kg, 5 mg/kg, 10 mg/kg, or 15 mg/kg. In some embodiments, the dose is 1 mg/kg, 2.5 mg/kg, or 5 mg/kg. In some embodiments, the dose is administered every about every two weeks or about every three weeks. In other embodiments, the dose is administered every week, every ten days, every four weeks, every six weeks or every two months.

[00173] In certain embodiments, the carboplatin dosage is 300-500 mg/m^2. The dosage may be based on the patient's glomular filtration rate and is generally described in terms of area under curve (AUC). Dosing may be AUC 4, AUC 5, or AUC 6, as determined the treating physician.

[00174] Paclitaxel dosing can be given at a dose of 175 mg/m^2, but also be in the range of 50-300 mg/m^2, for example, about 50, 75, 100, 125, 135, 150, 175, 200, 225, 250, or 300 mg/m^2.

[00175] In particular embodiments, the carboplatin and paclitaxel are given, e.g., by intravenous infusion administered, e.g., about every 3 weeks. In other embodiments, the drugs are administered about weekly, every other week, every four weeks, every six weeks, every eight weeks, or every 3 months.

VI. Additional combination therapy

[00176] In certain embodiments, in addition to administering a VEGF/DLL4-binding agent (e.g., an antibody) or therapeutic combination described herein, the method or treatment may further comprise administering at least one additional therapeutic agent. In some embodiments, the at least one additional therapeutic agent comprises 1, 2, 3, or more additional therapeutic agents.

[00177] Combination therapy with at least two therapeutic agents often uses agents that work by different mechanisms of action, although this is not required. Combination therapy using agents with different mechanisms of action may result in additive or synergetic effects. Combination therapy may allow for a lower dose of each agent than is used in monotherapy, thereby reducing toxic side effects and/or increasing the therapeutic index of at least one of the agents. Combination therapy may decrease the likelihood that resistant cancer cells will develop. In some embodiments, combination therapy comprises a therapeutic agent that primarily affects (e.g., inhibits or kills) non-tumorigenic cells and a therapeutic agent that primarily affects (e.g., inhibits or kills) tumorigenic CSCs.

[00178] Useful classes of therapeutic agents include, for example, antitubulin agents, auristatins, DNA minor groove binders, DNA replication inhibitors, alkylating agents (e.g., platinum complexes such as cisplatin, mono(platinum), bis(platinum) and tri-nuclear platinum complexes and carboplatin), anthracyclines, antibiotics, antifolates, antimetabolites, chemotherapy sensitizers, duocarmycins, etoposides, fluorinated pyrimidines, ionophores, lexitropsins, nitrosoureas, platinols, purine antimetabolites, puromycins, radiation sensitizers, steroids, taxanes, topoisomerase inhibitors, vinca alkaloids, or the like. In certain embodiments, the additional therapeutic agent is an alkylating agent, an antimetabolite, an antimitotic, a topoisomerase inhibitor, or an angiogenesis inhibitor.

[00179] Further therapeutic agents that may be administered with the VEGF/DLL4-binding agents or therapeutic combinations include chemotherapeutic agents. Thus, in some embodiments, the method or treatment involves the administration of an anti-VEGF-binding agent or antibody or therapeutic combination of the present invention in conjunction with a further chemotherapeutic agent or cocktail of multiple different chemotherapeutic agents. In some embodiments, the method or treatment involves the administration of an anti-DLL4-binding agent or antibody or therapeutic combination of the present invention in conjunction with a further chemotherapeutic agent or cocktail of multiple different chemotherapeutic agents. In some embodiments, the method or treatment involves the administration of a bispecific antibody of the present invention that binds VEGF and DLL4 or therapeutic combination in conjunction with a further chemotherapeutic agent or cocktail of multiple different chemotherapeutic agents.

[00180] Chemotherapeutic agents useful in the instant invention include, but are not limited to, alkylating agents such as thiotepa and cyclophosphamide (CYTOXAN); alkyl sulfonates such as busulfan, improsulfan and piposulfan; aziridines such as benzodopa, carboquone, meturedopa, and uredopa; ethylenimines and methylamelamines including altretamine, triethylenemelamine, trietylenephosphoramide, triethylenethiophosphaoramide and trimethylolomelamime; nitrogen mustards such as chlorambucil, chlornaphazine, cholophosphamide, estramustine, ifosfamide, mechlorethamine, mechlorethamine oxide hydrochloride, melphalan, novembichin, phenesterine, prednimustine, trofosfamide, uracil mustard; nitrosureas such as carmustine, chlorozotocin, fotemustine, lomustine,

nimustine, ranimustine; antibiotics such as aclacinomysins, actinomycin, authramycin, azaserine, bleomycins, cactinomycin, calicheamicin, carabicin, caminomycin, carzinophilin, chromomycins, dactinomycin, daunorubicin, detorubicin, 6-diazo-5-oxo-L-norleucine, doxorubicin (adriamycin), epirubicin, esorubicin, idarubicin, marcellomycin, mitomycins, mycophenolic acid, nogalamycin, olivomycins, peplomycin, potfiromycin, puromycin, quelamycin, rodorubicin, streptonigrin, streptozocin, tubercidin, ubenimex, zinostatin, zorubicin; anti-metabolites such as methotrexate and 5-fluorouracil (5-FU); folic acid analogues such as denopterin, methotrexate, pteropterin, trimetrexate; purine analogs such as fludarabine, 6-mercaptopurine, thiamiprine, thioguanine; pyrimidine analogs such as ancitabine, azacitidine, 6-azauridine, carmofur, cytosine arabinoside, dideoxyuridine, doxifluridine, enocitabine, floxuridine, 5-FU; androgens such as calusterone, dromostanolone propionate, epitiostanol, mepitiostane, testolactone; anti-adrenals such as aminoglutethimide, mitotane, trilostane; folic acid replenishers such as folinic acid; aceglatone; aldophosphamide glycoside; aminolevulinic acid; amsacrine; bestrabucil; bisantrene; edatraxate; defofamine; demecolcine; diaziquone; elformithine; elliptinium acetate; etoglucid; gallium nitrate; hydroxyurea; lentinan; lonidamine; mitoguazone; mitoxantrone; mopidamol; nitracrine; pentostatin; phenamet; pirarubicin; podophyllinic acid; 2-ethylhydrazide; procarbazine; PSK; razoxane; sizofuran; spirogermanium; tenuazonic acid; triaziquone; 2,2',2''-trichlorotriethylamine; urethan; vindesine; dacarbazine; mannomustine; mitobronitol; mitolactol; pipobroman; gacytosine; arabinoside (Ara-C); taxoids, e.g. paclitaxel (TAXOL) and docetaxel (TAXOTERE); chlorambucil; gemcitabine; 6-thioguanine; mercaptopurine; platinum analogs such as cisplatin, carboplatin, oxaliplatin; vinblastine; platinum; etoposide (VP-16); ifosfamide; mitomycin C; mitoxantrone; vincristine; vinorelbine; navelbine; novantrone; teniposide; daunomycin; aminopterin; ibandronate; CPT11; topoisomerase inhibitor RFS 2000; difluoromethylornithine (DMFO); retinoic acid; esperamicins; capecitabine (XELODA); and pharmaceutically acceptable salts, acids or derivatives of any of the above. Chemotherapeutic agents also include anti-hormonal agents that act to regulate or inhibit hormone action on tumors such as anti-estrogens including, for example, tamoxifen, raloxifene, aromatase inhibiting 4(5)-imidazoles, 4-hydroxytamoxifen, trioxifene, keoxifene, LY117018, onapristone, and toremifene (FARESTON); and anti-androgens such as flutamide, nilutamide, bicalutamide, leuprolide, and goserelin; and pharmaceutically acceptable salts, acids or derivatives of any of the above. In certain embodiments, an additional therapeutic agent is cisplatin. In certain embodiments, an additional therapeutic agent is oxaliplatin. In some embodiments, an additional agent is doxorubicin (adriamycin). In some embodiments, an additional agent is epirubicin.

[00181] In certain embodiments, the chemotherapeutic agent is a topoisomerase inhibitor. Topoisomerase inhibitors are chemotherapeutic agents that interfere with the action of a topoisomerase enzyme (e.g., topoisomerase I or II). Topoisomerase inhibitors include, but are not limited to, doxorubicin HCl, daunorubicin citrate, mitoxantrone HCl, actinomycin D, etoposide, topotecan HCl, teniposide (VM-26),

and irinotecan, as well as pharmaceutically acceptable salts, acids, or derivatives of any of these. In certain embodiments, an additional therapeutic agent is irinotecan.

[00182] In certain embodiments, the chemotherapeutic agent is an anti-metabolite. An anti-metabolite is a chemical with a structure that is similar to a metabolite required for normal biochemical reactions, yet different enough to interfere with one or more normal functions of cells, such as cell division. Anti-metabolites include, but are not limited to, gemcitabine, fluorouracil, capecitabine, methotrexate sodium, ralitrexed, pemetrexed, tegafur, cytosine arabinoside, thioguanine, 5-azacytidine, 6-mercaptopurine, azathioprine, 6-thioguanine, pentostatin, fludarabine phosphate, and cladribine, as well as pharmaceutically acceptable salts, acids, or derivatives of any of these. In some embodiments, an additional therapeutic agent is 5-fluorouracil. In some embodiments, additional agents are 5-fluorouracil and irinotecan. In some embodiments, additional agents are 5-fluorouracil and oxaliplatin. In some embodiments, additional agents are 5-fluorouracil and cisplatin. In some embodiments, an additional agent is methotrexate.

[00183] In certain embodiments, the chemotherapeutic agent is an antimitotic agent, including, but not limited to, agents that bind tubulin. In some embodiments, the agent is a taxane. In certain embodiments, the agent is paclitaxel or docetaxel, or a pharmaceutically acceptable salt, acid, or derivative of paclitaxel or docetaxel. In certain embodiments, the agent is paclitaxel (TAXOL), docetaxel (TAXOTERE), albumin-bound paclitaxel (ABRAXANE®), DHA-paclitaxel, or PG-paclitaxel. In some embodiments, the antimitotic agent comprises a vinca alkaloid, such as vincristine, binblastine, vinorelbine, or vindesine, or pharmaceutically acceptable salts, acids, or derivatives thereof. In some embodiments, the antimitotic agent is an inhibitor of kinesin Eg5 or an inhibitor of a mitotic kinase such as Aurora A or Plk1. In some embodiments, an additional agent is docetaxel.

[00184] In some embodiments, an additional therapeutic agent comprises an agent such as a small molecule. For example, treatment can involve the combined administration of a VEGF/DLL4-binding agent (e.g. an antibody) or therapeutic combination of the present invention with a small molecule that acts as an inhibitor against additional tumor-associated proteins including, but not limited to, EGFR, ErbB2, HER2, and/or VEGF. In certain embodiments, the additional therapeutic agent is a small molecule that inhibits a cancer stem cell pathway. In some embodiments, the additional therapeutic agent is a small molecule inhibitor of the Notch pathway. In some embodiments, the additional therapeutic agent is a small molecule inhibitor of the Wnt pathway. In some embodiments, the additional therapeutic agent is a small molecule inhibitor of the BMP pathway. In some embodiments, the additional therapeutic agent is a small molecule that inhibits β-catenin signaling.

[00185] In some embodiments, the further therapeutic agent comprises a biological molecule, such as an antibody. For example, treatment can involve the combined administration of a VEGF/DLL4-binding agent (e.g. an antibody) or therapeutic combination of the present invention with further antibodies against

additional tumor-associated proteins including, but not limited to, antibodies that bind EGFR, ErbB2, HER2, VEGF and/or VEGF receptors. In some embodiments, the additional therapeutic agent is anti-HER2 antibody trastuzumab. In some embodiments, the additional therapeutic agent is anti-VEGFR-2 antibody ramucirumab. In certain embodiments, the additional therapeutic agent is an antibody that is an anti-cancer stem cell marker antibody. In some embodiments, the additional therapeutic agent is an antibody that binds a component of the Notch pathway. In some embodiments, the additional therapeutic agent is an antibody that binds a component of the Wnt pathway. In certain embodiments, the additional therapeutic agent is an antibody that inhibits a cancer stem cell pathway. In some embodiments, the additional therapeutic agent is an antibody inhibitor of the Notch pathway. In some embodiments, the additional therapeutic agent is an antibody inhibitor of the Wnt pathway. In some embodiments, the additional therapeutic agent is an antibody inhibitor of the BMP pathway. In some embodiments, the additional therapeutic agent is an antibody that inhibits β-catenin signaling. In certain embodiments, the additional therapeutic agent is an antibody that is an angiogenesis inhibitor or modulator (e.g., an anti-VEGF or VEGF receptor antibody). In certain embodiments, the additional therapeutic agent is bevacizumab (AVASTIN), trastuzumab (HERCEPTIN), panitumumab (VECTIBIX), or cetuximab (ERBITUX).

[00186] Furthermore, treatment with a VEGF/DLL4-binding agent or therapeutic combination described herein can include further treatment with other biologic molecules, such as one or more cytokines (e.g., lymphokines, interleukins, tumor necrosis factors, and/or growth factors) or can be accompanied by surgical removal of tumors, cancer cells, or any other therapy deemed necessary by a treating physician.

[00187] It will be appreciated that the VEGF/DLL4-binding agent or therapeutic combination and an additional therapeutic agent may be administered in any order or concurrently. In some embodiments, treatment with a VEGF/DLL4-binding agent (e.g., an antibody) can occur prior to, concurrently with, or subsequent to administration of chemotherapies. Combined administration may include co-administration, either in a single pharmaceutical formulation or using separate formulations, or consecutive administration in either order but generally within a time period such that all active agents can exert their biological activities simultaneously. Preparation and dosing schedules for such chemotherapeutic agents can be used according to manufacturers' instructions or as determined empirically by the skilled practitioner. Preparation and dosing schedules for such chemotherapy are also described in *The Chemotherapy Source Book, 4th Edition*, 2008, M. C. Perry, Editor, Lippincott, Williams & Wilkins, Philadelphia, PA.

[00188] In certain embodiments, the VEGF/DLL4-binding agent or therapeutic combination and an additional therapeutic agent will be administered substantially simultaneously or concurrently. For example, a subject may be given a VEGF/DLL4-binding agent (e.g., an antibody) or therapeutic combination while undergoing a course of treatment with an additional therapeutic agent (e.g.,

chemotherapy). In certain embodiments, a VEGF/DLL4-binding agent will be administered within 1 year of the treatment with an additional therapeutic agent. In certain alternative embodiments, a VEGF/DLL4-binding agent will be administered within 10, 8, 6, 4, or 2 months of any treatment with an additional therapeutic agent. In certain other embodiments, a VEGF/DLL4-binding agent will be administered within 4, 3, 2, or 1 weeks of any treatment with an additional therapeutic agent. In some embodiments, a VEGF/DLL4-binding agent will be administered within 5, 4, 3, 2, or 1 days of any treatment with an additional therapeutic agent. It will further be appreciated that the agents or treatments may be administered to the subject within a matter of hours or minutes (i.e., substantially simultaneously).

[00189] In certain embodiments, the treatment of cancer involves the administration of a VEGF/DLL4-binding agent (e.g. an antibody) or therapeutic combination of the present invention in combination with radiation therapy. Treatment with a VEGF/DLL4-binding agent or therapeutic combination can occur prior to, concurrently with, or subsequent to administration of radiation therapy. Dosing schedules for such radiation therapy can be determined by the skilled medical practitioner.

[00190] In certain embodiments, the treatment of cancer involves the administration of a VEGF/DLL4-binding agent (e.g. an antibody) or therapeutic combination of the present invention in combination with a surgical procedure. Treatment with a VEGF/DLL4-binding agent or therapeutic combination can occur prior to, concurrently with, or subsequent to the surgical procedure.

[00191] For the treatment of cancer, the appropriate dosage of an VEGF/DLL4-binding agent (e.g., an antibody) or therapeutic combination of the present invention depends on the severity and course of the cancer, the responsiveness of the cancer, whether the VEGF/DLL4-binding agent or antibody or therapeutic combination is administered for therapeutic or preventative purposes, previous therapy the patient has received, the patient's clinical history, and so on, all at the discretion of the treating physician. The VEGF/DLL4-binding agent or antibody or therapeutic combination can be administered one time or as a series of treatments spread over several days to several months, or until a cure is effected or a diminution of the disease state is achieved (e.g., reduction in tumor size). Optimal dosing schedules can be calculated from measurements of drug accumulation in the body of the patient and will vary depending on the relative potency of an individual antibody or agent. The administering physician can determine optimum dosages, dosing methodologies, and repetition rates.

[00192] In certain embodiments, dosage of a VEGF/DLL4-binding agent or antibody is from about 0.01μg to about 100mg/kg of body weight, from about 0.1μg to about 100mg/kg of body weight, from about 1μg to about 100mg/kg of body weight, from about 1mg to about 100mg/kg of body weight, about 1mg to about 80mg/kg of body weight from about 10mg to about 100mg/kg of body weight, from about 10mg to about 75mg/kg of body weight, or from about 10mg to about 50mg/kg of body weight. In certain embodiments, the dosage of the antibody or other VEGF/DLL4-binding agent is from about 0.1mg to about 20mg/kg of body weight. In certain embodiments, dosage can be given once or more daily, weekly,

monthly, or yearly. In certain embodiments, the antibody or other VEGF/DLL4-binding agent is given once every week, once every two weeks, once every three weeks, or once every month.

[00193] In some embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) may be administered at an initial higher "loading" dose, followed by one or more lower doses. In some embodiments, the frequency of administration may also change. In some embodiments, a dosing regimen may comprise administering an initial dose, followed by additional doses (or "maintenance" doses) once a week, once every two weeks, once every three weeks, or once every month. For example, a dosing regimen may comprise administering an initial loading dose, followed by a weekly maintenance dose of, for example, one-half of the initial dose. Or a dosing regimen may comprise administering an initial loading dose, followed by maintenance doses of, for example one-half of the initial dose every other week. Or a dosing regimen may comprise administering three initial doses for 3 weeks, followed by maintenance doses of, for example, the same amount every other week. Or a dosing regimen may comprise administering an initial dose followed by additional doses every 3 weeks or once a month. The treating physician can estimate repetition rates for dosing based on measured residence times and concentrations of the drug in bodily fluids or tissues. The progress of therapy can be monitored by conventional techniques and assays.

[00194] As is known to those of skill in the art, administration of any therapeutic agent may lead to side effects and/or toxicities. In some cases, the side effects and/or toxicities are so severe as to preclude administration of the particular agent at a therapeutically effective dose. In some cases, drug therapy must be discontinued, and other agents may be tried. However, many agents in the same therapeutic class often display similar side effects and/or toxicities, meaning that the patient either has to stop therapy, or if possible, suffer from the unpleasant side effects associated with the therapeutic agent.

[00195] Side effects from therapeutic agents may include, but are not limited to, hives, skin rashes, itching, nausea, vomiting, decreased appetite, diarrhea, chills, fever, fatigue, muscle aches and pain, headaches, low blood pressure, high blood pressure, hypokalemia, low blood counts, bleeding, and cardiac problems.

[00196] Thus, one aspect of the present invention is directed to methods of treating cancer (e.g., colorectal, ovarian, pancreatic, or endometrial cancer) in a patient comprising administering an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination using an intermittent dosing regimen, which may reduce side effects and/or toxicities associated with administration of the anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination. As used herein, "intermittent dosing" refers to a dosing regimen using a dosing interval of more than once a week, e.g., dosing once every 2 weeks, once every 3 weeks, once every 4 weeks, etc. In some embodiments, a method for treating cancer in a human patient comprises administering to the patient an effective dose of an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination according to an intermittent dosing regimen. In some embodiments, a method for treating cancer in a human patient comprises administering to the patient an effective dose of

an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination according to an intermittent dosing regimen, and increasing the therapeutic index of the anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination. In some embodiments, the intermittent dosing regimen comprises administering an initial dose of an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination to the patient, and administering subsequent doses of the anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination about once every 2 weeks. In some embodiments, the intermittent dosing regimen comprises administering an initial dose of an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination to the patient, and administering subsequent doses of the anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination about once every 3 weeks. In some embodiments, the intermittent dosing regimen comprises administering an initial dose of an anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination to the patient, and administering subsequent doses of the anti-VEGF/anti-DLL4 bispecific antibody or therapeutic combination about once every 4 weeks.

[00197] In some embodiments, the subsequent doses in an intermittent dosing regimen are about the same amount or less than the initial dose. In other embodiments, the subsequent doses are a greater amount than the initial dose. As is known by those of skill in the art, doses used will vary depending on the clinical goals to be achieved. In some embodiments, the initial dose is about 0.25mg/kg to about 20mg/kg. In some embodiments, the initial dose is about 0.5, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20mg/kg. In certain embodiments, the initial dose is about 0.5mg/kg. In certain embodiments, the initial dose is about 1mg/kg. In certain embodiments, the initial dose is about 2.5mg/kg. In certain embodiments, the initial dose is about 5mg/kg. In certain embodiments, the initial dose is about 7.5mg/kg. In certain embodiments, the initial dose is about 10mg/kg. In certain embodiments, the initial dose is about 12.5mg/kg. In certain embodiments, the initial dose is about 15mg/kg. In certain embodiments, the initial dose is about 20mg/kg. In some embodiments, the subsequent doses are about 0.25mg/kg to about 15mg/kg. In certain embodiments, the subsequent doses are about 0.5, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15mg/kg. In certain embodiments, the subsequent doses are about 0.5mg/kg. In certain embodiments, the subsequent doses are about 1mg/kg. In certain embodiments, the subsequent doses are about 2.5mg/kg. In certain embodiments, the subsequent doses are about 5mg/kg. In some embodiments, the subsequent doses are about 7.5mg/kg. In some embodiments, the subsequent doses are about 10mg/kg. In some embodiments, the subsequent doses are about 12.5mg/kg.

[00198] In some embodiments, a dosing regimen may be limited to a specific number of administrations or "cycles". In some embodiments, the antibodies described herein are administered for 3, 4, 5, 6, 7, 8, or more cycles. In some embodiments, the antibodies described herein are administered for 3, 4, 5, 6, 7, 8, or more cycles in combination with intermittent dosing. For example, an antibody is administered every 3 weeks for 6 cycles, an antibody is administered every 4 weeks for 6 cycles, an antibody is administered

every 3 weeks for 4 cycles, an antibody is administered every 4 weeks for 4 cycles, etc. Dosing schedules can be decided upon and subsequently modified by those skilled in the art.

[00199] The choice of delivery method for the initial and subsequent doses is made according to the ability of the animal or human patient to tolerate introduction of the anti-VEGF/anti-DLL4 bispecific antibody into the body. Thus, in any of the aspects and/or embodiments described herein, the administration of the anti-VEGF/anti-DLL4 bispecific antibody may be by intravenous injection or intravenously. In some embodiments, the administration is by intravenous infusion. In any of the aspects and/or embodiments described herein, the administration of the anti-VEGF/anti-DLL4 bispecific antibody may be by a non-intravenous route.

III. Antibodies

[00200] The methods, compositions, and kits described herein include agents that specifically bind human VEGF proteins and/or human DLL4 proteins. These agents are referred to herein as "VEGF/DLL4-binding agents". The phrase "VEGF/DLL4-binding agent" encompasses agents that bind only VEGF, agents that bind only DLL4, and bispecific agents that bind both VEGF and DLL4. In certain embodiments, in addition to specifically binding VEGF and/or DLL4, the VEGF/DLL4-binding agents further specifically bind at least one additional target or antigen. In some embodiments, the VEGF/DLL4-binding agent is an antibody. In some embodiments, the VEGF/DLL4-binding agent is a polypeptide. In certain embodiments, the VEGF/DLL4-binding agent specifically binds human VEGF. In certain embodiments, the VEGF/DLL4-binding agent specifically binds human DLL4. In certain embodiments, the VEGF/DLL4-binding agent is a bispecific antibody, including molecules such as dual variable domain immunoglobulins (DVD-Igs; see, e.g., Jakob et al., *MAbs* 5:358-63, 2013). In certain embodiments, the VEGF/DLL4-binding agent is a bispecific antibody that specifically binds human VEGF and human DLL4. The full-length amino acid (aa) sequences for human VEGF (VEGF-A) and human DLL4 are known in the art and are provided herein as SEQ ID NO:27 (VEGF) and SEQ ID NO:23 (DLL4).

[00201] In certain embodiments, the VEGF/DLL4-binding agent or antibody binds VEGF and/or DLL4 with a dissociation constant (K_D) of about 1μM or less, about 100nM or less, about 40nM or less, about 20nM or less, about 10nM or less, about 1nM or less, or about 0.1nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds VEGF and/or DLL4 with a K_D of about 20nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds VEGF and/or DLL4 with a K_D of about 10nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds VEGF and/or DLL4 with a K_D of about 1nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds VEGF and/or DLL4 with a K_D of about 0.1nM or less. In some embodiments, the VEGF/DLL4-binding agent binds both human VEGF and mouse VEGF with a K_D of about 100nM or less. In some embodiments, the VEGF/DLL4-binding agent binds both human VEGF and mouse VEGF with a K_D of

about 50nM or less. In some embodiments, a VEGF/DLL4-binding agent binds both human DLL4 and mouse DLL4 with a K_D of about 100nM or less. In some embodiments, a VEGF/DLL4-binding agent binds both human DLL4 and mouse DLL4 with a K_D of about 50nM or less. In some embodiments, the dissociation constant of the binding agent (e.g., an antibody) to VEGF is the dissociation constant determined using a VEGF fusion protein comprising at least a portion of VEGF immobilized on a Biacore chip. In some embodiments, the dissociation constant of the binding agent (e.g., an antibody) to DLL4 is the dissociation constant determined using a DLL4-fusion protein comprising at least a portion of DLL4 immobilized on a Biacore chip.

[00202] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first antigen-binding site that specifically binds VEGF and a second antigen-binding site that specifically binds DLL4. In some embodiments, a VEGF/DLL4-binding agent or antibody binds both VEGF and DLL4 with a K_D of about 100nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds both VEGF and DLL4 with a K_D of about 50nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds both VEGF and DLL4 with a K_D of about 20nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds both VEGF and DLL4 with a K_D of about 10nM or less. In some embodiments, a VEGF/DLL4-binding agent or antibody binds both VEGF and DLL4 with a K_D of about 1nM or less. In some embodiments, the affinity of one of the antigen-binding sites may be weaker than the affinity of the other antigen-binding site. For example, the K_D of one antigen binding site may be about 1nM and the K_D of the second antigen-binding site may be about 10nM. In some embodiments, the difference in affinity between the two antigen-binding sites may be about 2-fold or more, about 3-fold or more, about 5-fold or more, about 8-fold or more, about 10-fold or more, about 15-fold or more, about 20-fold or more, about 30-fold or more, about 50-fold or more, or about 100-fold or more. Modulation of the affinities of the two antigen-binding sites may affect the biological activity of the bispecific antibody. For example, decreasing the affinity of the antigen-binding site for DLL4 or VEGF, may have a desirable effect, for example decreased toxicity of the binding agent or increased therapeutic index.

[00203] By way of non-limiting example, the bispecific antibody may comprise (a) a first antigen-binding site that binds human VEGF with a K_D between about 0.1nM and about 1.0nM, and (b) a second antigen-binding site that specifically binds human DLL4 with a K_D between about 0.1nM and about 20nM, between about 0.5nM and about 20nM, between about 1.0nM and 10nM. In certain embodiments, the bispecific antibody comprises two identical light chains.

[00204] In certain embodiments, the VEGF/DLL4-binding agent (e.g., an antibody) binds VEGF and/or DLL4 with a half maximal effective concentration (EC_{50}) of about 1μM or less, about 100nM or less, about 40nM or less, about 20nM or less, about 10nM or less, about 1nM or less, or about 0.1nM or less. In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) binds VEGF and/or DLL4 with

a half maximal effective concentration (EC_{50}) of about 1μM or less, about 100nM or less, about 40nM or less, about 20nM or less, about 10nM or less, about 1nM or less, or about 0.1nM or less.

[00205] In certain embodiments, the VEGF/DLL4-binding agent is an antibody. In some embodiments, the antibody is a recombinant antibody. In some embodiments, the antibody is a monoclonal antibody. In some embodiments, the antibody is a chimeric antibody. In some embodiments, the antibody is a humanized antibody. In some embodiments, the antibody is a human antibody. In certain embodiments, the antibody is an IgA, IgD, IgE, IgG, or IgM antibody. In certain embodiments, the antibody is an IgG1 antibody. In certain embodiments, the antibody is an IgG2 antibody. In certain embodiments, the antibody is an antibody fragment comprising an antigen-binding site. In some embodiments, the antibody is a bispecific antibody. In some embodiments, the antibody is monovalent, monospecific, bivalent, or multispecific. In some embodiments, the antibody is conjugated to a cytotoxic moiety. In some embodiments, the antibody is isolated. In some embodiments, the antibody is substantially pure.

[00206] The VEGF/DLL4-binding agents (e.g., antibodies) of the present invention can be assayed for specific binding by any method known in the art. The immunoassays which can be used include, but are not limited to, competitive and non-competitive assay systems using techniques such as Biacore analysis, FACS analysis, immunofluorescence, immunocytochemistry, Western blot analysis, radioimmunoassay, ELISA, "sandwich" immunoassay, immunoprecipitation assay, precipitation reaction, gel diffusion precipitin reaction, immunodiffusion assay, agglutination assay, complement-fixation assay, immunoradiometric assay, fluorescent immunoassay, homogeneous time-resolved fluorescence assay (HTRF), and protein A immunoassay. Such assays are routine and well-known in the art (see, e.g., Ausubel et al., Editors, 1994-present, *Current Protocols in Molecular Biology*, John Wiley & Sons, Inc., New York, NY).

[00207] For example, the specific binding of an antibody to human VEGF and/or human DLL4 may be determined using ELISA. An ELISA assay comprises preparing antigen, coating wells of a 96 well microtiter plate with antigen, adding the antibody or other binding agent conjugated to a detectable compound such as an enzymatic substrate (e.g. horseradish peroxidase or alkaline phosphatase) to the well, incubating for a period of time, and detecting the presence of the binding agent bound to the antigen. In some embodiments, the binding agent or antibody is not conjugated to a detectable compound, but instead a second antibody that recognizes the binding agent or antibody (e.g., an anti-Fc antibody) and is conjugated to a detectable compound is added to the well. In some embodiments, instead of coating the well with the antigen, the binding agent or antibody can be coated to the well and a second antibody conjugated to a detectable compound can be added following the addition of the antigen to the coated well. One of skill in the art would be knowledgeable as to the parameters that can be modified to increase the signal detected as well as other variations of ELISAs known in the art.

[00208] In another example, the specific binding of an antibody to human VEGF and/or human DLL4 may be determined using FACS. A FACS screening assay may comprise generating a cDNA construct that expresses an antigen as a fusion protein, transfecting the construct into cells, expressing the antigen on the surface of the cells, mixing the binding agent or antibody with the transfected cells, and incubating for a period of time. The cells bound by the binding agent or antibody may be identified by using a secondary antibody conjugated to a detectable compound (e.g., PE-conjugated anti-Fc antibody) and a flow cytometer. One of skill in the art would be knowledgeable as to the parameters that can be modified to optimize the signal detected as well as other variations of FACS that may enhance screening (e.g., screening for blocking antibodies).

[00209] The binding affinity of an antibody or other binding-agent to an antigen (e.g., VEGF or DLL4) and the off-rate of an antibody-antigen interaction can be determined by competitive binding assays. One example of a competitive binding assay is a radioimmunoassay comprising the incubation of labeled antigen (e.g., ^{3}H or ^{125}I), or fragment or variant thereof, with the antibody of interest in the presence of increasing amounts of unlabeled antigen followed by the detection of the antibody bound to the labeled antigen. The affinity of the antibody for the antigen and the binding off-rates can be determined from the data by Scatchard plot analysis. In some embodiments, Biacore kinetic analysis is used to determine the binding on and off rates of antibodies or agents that bind an antigen (e.g., VEGF or DLL4). Biacore kinetic analysis comprises analyzing the binding and dissociation of antibodies from chips with immobilized antigen (e.g., VEGF or DLL4) on their surface.

[00210] In certain embodiments, the invention provides a VEGF-binding agent (e.g., an antibody) that specifically binds human VEGF, wherein the VEGF-binding agent (e.g., an antibody) comprises one, two, three, four, five, and/or six of the CDRs of antibody 219R45 (see Table 1). In some embodiments, the VEGF-binding agent comprises one or more of the CDRs of 219R45, two or more of the CDRs of 219R45, three or more of the CDRs of 219R45, four or more of the CDRs of 219R45, five or more of the CDRs of 219R45, or all six of the CDRs of 219R45. In some embodiments, the VEGF-binding agent binds human VEGF and mouse VEGF.

Table 1

	219R45
HC CDR1	NYWMH (SEQ ID NO:17)
HC CDR2	DINPSNGRTSYKEKFKR (SEQ ID NO:18)
HC CDR3	HYDDKYYPLMDY (SEQ ID NO:19)
LC CDR1	RASESVDNYGISFMK (SEQ ID NO:20)

LC CDR2	AASNQGS (SEQ ID NO:21)
LC CDR3	QQSKEVPWTFGG (SEQ ID NO:22)

[00211] In certain embodiments, the invention provides a VEGF-binding agent (e.g., an antibody) that specifically binds human VEGF, wherein the VEGF-binding agent comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19). In some embodiments, the VEGF-binding agent further comprises a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In certain embodiments, the VEGF-binding agent comprises: (a) a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00212] In certain embodiments, the invention provides a VEGF-binding agent (e.g., an antibody) that specifically binds human VEGF, wherein the VEGF-binding agent comprises: (a) a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (b) a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (c) a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (d) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (e) a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; and (f) a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions. In certain embodiments, the amino acid substitutions are conservative substitutions.

[00213] In certain embodiments, the invention provides a VEGF-binding agent (e.g., an antibody) that specifically binds VEGF, wherein the VEGF-binding agent comprises a heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:11, and a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the VEGF-binding agent comprises a heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:11. In certain embodiments, the VEGF-binding agent comprises a light chain variable region having at least about 85%, at least about

90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12. In certain embodiments, the VEGF-binding agent comprises a heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11, and a light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the VEGF-binding agent comprises a heavy chain variable region comprising SEQ ID NO:11, and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, the VEGF-binding agent comprises a heavy chain variable region consisting essentially of SEQ ID NO:11, and a light chain variable region consisting essentially of SEQ ID NO:12. In some embodiments, the VEGF-binding agent comprises a heavy chain comprising SEQ ID NO:49, and a light chain comprising SEQ ID NO:8. In some embodiments, the VEGF-binding antibody or other agent comprises a heavy chain comprising SEQ IDNO:7, and a light chain comprising SEQ ID NO:8.

[00214] In some embodiments, the VEGF-binding agent binds VEGF with a K_D of about 10nM or less. In some embodiments, the VEGF-binding agent binds VEGF with a K_D of about 1nM or less. In some embodiments, the VEGF-binding agent binds VEGF with a K_D of about 0.1nM or less. In some embodiments, the VEGF-binding agent binds VEGF with a K_D of about 0.01nM or less. In some embodiments, at least one amino acid residue in at least one CDR of the VEGF-binding agent is substituted with a different amino acid so that the affinity of the VEGF-binding agent for VEGF is altered. In some embodiments, the affinity of the VEGF-binding agent is increased. In some embodiments, the affinity of the VEGF-binding agent is decreased. In some embodiments, the VEGF-binding agent binds human VEGF. In some embodiments, the VEGF-binding agent binds human VEGF and mouse VEGF.

[00215] In certain embodiments, the VEGF-binding agent comprises the heavy chain variable region and light chain variable region of the 219R45 antibody. In certain embodiments, the VEGF-binding agent comprises the heavy chain and light chain of the 219R45 antibody (with or without the leader sequence). In certain embodiments, a VEGF-binding agent is the 219R45 antibody. In some embodiments, the VEGF-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13236. The plasmid PTA-13236 was deposited with the American Type Culture Collection (ATCC), at 10801 University Boulevard, Manassas, VA, 20110, under the conditions of the Budapest Treaty on September 21, 2012. In some embodiments, the VEGF-binding agent comprises the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235. The plasmid PTA-13235 was deposited with the ATCC, at 10801 University Boulevard, Manassas, VA, 20110, under the conditions of the Budapest Treaty on September 21, 2012. In some embodiments, the VEGF-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13236 and the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235.

[00216] In certain embodiments, a VEGF-binding agent comprises, consists essentially of, or consists of, the antibody 219R45.

[00217] In certain embodiments, a VEGF-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on VEGF as an antibody of the invention. In another embodiment, a VEGF-binding agent is an antibody that binds an epitope on VEGF that overlaps with the epitope on VEGF bound by an antibody of the invention. In certain embodiments, a VEGF-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on VEGF as antibody 219R45. In another embodiment, the VEGF-binding agent is an antibody that binds an epitope on VEGF that overlaps with the epitope on VEGF bound by antibody 219R45.

[00218] In some embodiments, the VEGF-binding agent inhibits binding of VEGF to at least one VEGF receptor. In certain embodiments, the VEGF-binding agent inhibits binding of human VEGF to VEGFR-1 or VEGFR-2. In some embodiments, the VEGF-binding agent specifically binds VEGF and modulates angiogenesis. In some embodiments, the VEGF-binding agent specifically binds VEGF and inhibits angiogenesis. In some embodiments, the VEGF-binding agent specifically binds VEGF and inhibits tumor growth.

[00219] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent (e.g., an antibody) comprises one, two, three, four, five, and/or six of the CDRs of antibody 21R79 (see Table 2). In some embodiments, the DLL4-binding agent comprises one or more of the CDRs of 21R79, two or more of the CDRs of 21R79, three or more of the CDRs of 21R79, four or more of the CDRs of 21R79, five or more of the CDRs of 21R79, or all six of the CDRs of 21R79. In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent (e.g., an antibody) comprises one, two, three, four, five, and/or six of the CDRs of antibody 21R75 (see Table 2). In some embodiments, the DLL4-binding agent comprises one or more of the CDRs of 21R75, two or more of the CDRs of 21R75, three or more of the CDRs of 21R75, four or more of the CDRs of 21R75, five or more of the CDRs of 21R75, or all six of the CDRs of 21R75. In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent (e.g., an antibody) comprises one, two, three, four, five, and/or six of the CDRs of antibody 21R83 (see Table 2). In some embodiments, the DLL4-binding agent comprises one or more of the CDRs of 21R83, two or more of the CDRs of 21R83, three or more of the CDRs of 21R83, four or more of the CDRs of 21R83, five or more of the CDRs of 21R83, or all six of the CDRs of 21R83. In some embodiments, the DLL4-binding agent binds human DLL4 and mouse DLL4.

Table 2

	21R79	21R75	21R83
HC CDR1	TAYYIH (SEQ ID NO:13)	TAYYIH (SEQ ID NO:13)	TAYYIH (SEQ ID NO:13)
HC CDR2	YIANYNRATNYNQKFKG (SEQ ID NO:14)	YIAGYKDATNYNQKFKG (SEQ ID NO:59)	YISNYNRATNYNQKFKG (SEQ ID NO:65)
HC CDR3	RDYDYDVGMDY (SEQ ID NO:16)	RDYDYDVGMDY (SEQ ID NO:16)	RDYDYDVGMDY (SEQ ID NO:16)
LC CDR1	RASESVDNYGISFMK (SEQ ID NO:20)	RASESVDNYGISFMK (SEQ ID NO:20)	RASESVDNYGISFMK (SEQ ID NO:20)
LC CDR2	AASNQGS (SEQ ID NO:21)	AASNQGS (SEQ ID NO:21)	AASNQGS (SEQ ID NO:21)
LC CDR3	QQSKEVPWTFGG (SEQ ID NO:22)	QQSKEVPWTFGG (SEQ ID NO:22)	QQSKEVPWTFGG (SEQ ID NO:22)

[00220] In certain embodiments, the heavy chain CDR1 of the DLL4-binding antibody is a minimal HC CDR1 comprising AYYIH (SEQ ID NO:79).

[00221] In some embodiments, the binding agent is an antibody that binds human DLL4 and comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00222] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the DLL4-binding agent further comprises a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In certain embodiments, the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00223] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (b) a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (c) a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (d) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (e) a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; and (f) a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions. In certain embodiments, the amino acid substitutions are conservative substitutions.

[00224] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds DLL4, wherein the DLL4-binding agent comprises a heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:10, and a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:10. In certain embodiments, the DLL4-binding agent comprises a light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:10, and a light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region comprising SEQ ID NO:10, and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region consisting essentially of SEQ ID NO:10, and a light chain variable region consisting essentially of SEQ ID NO:12. In some embodiments, the DLL4-binding agent comprises a heavy chain comprising SEQ ID NO:48, and a light chain comprising SEQ ID NO:8. In some embodiments, the DLL4-binding antibody or other agent comprises a heavy chain comprising SEQ ID NO:6, and a light chain comprising SEQ ID NO:8. In some embodiments, the antibody is a bispecific antibody.

[00225] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the DLL4-binding agent further comprises a light chain CDR1 comprising

RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In certain embodiments, the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00226] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (b) a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (c) a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (d) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (e) a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; and (f) a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions. In certain embodiments, the amino acid substitutions are conservative substitutions.

[00227] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds DLL4, wherein the DLL4-binding agent comprises a heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:58, and a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:58. In certain embodiments, the DLL4-binding agent comprises a light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:58, and a light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region comprising SEQ ID NO:58, and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region consisting essentially of SEQ ID NO:58, and a light chain variable region consisting essentially of SEQ ID NO:12. In some embodiments, the DLL4-binding agent comprises a heavy chain comprising SEQ ID NO:56, and a light chain comprising SEQ ID NO:8.

[00228] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the DLL4-binding agent further comprises a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In certain embodiments, the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00229] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds human DLL4, wherein the DLL4-binding agent comprises: (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (b) a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (c) a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (d) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (e) a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; and (f) a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions. In certain embodiments, the amino acid substitutions are conservative substitutions.

[00230] In certain embodiments, the invention provides a DLL4-binding agent (e.g., an antibody) that specifically binds DLL4, wherein the DLL4-binding agent comprises a heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:64, and a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:64. In certain embodiments, the DLL4-binding agent comprises a light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:64, and a light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a

heavy chain variable region comprising SEQ ID NO:64, and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, the DLL4-binding agent comprises a heavy chain variable region consisting essentially of SEQ ID NO:64, and a light chain variable region consisting essentially of SEQ ID NO:12. In some embodiments, the DLL4-binding agent comprises a heavy chain comprising SEQ ID NO:62, and a light chain comprising SEQ ID NO:8.In some embodiments, the agent is a bispecific antibody.

[00231] In some embodiments, the DLL4-binding agent is an antibody that comprises a heavy chain comprising SEQ ID NO:5, and a light chain comprising SEQ ID NO:8. In some embodiments, the antibody is a bispecific antibody.

[00232] In some embodiments, the DLL4-binding agent binds DLL4 with a K_D of 25nM or less. In some embodiments, the DLL4-binding agent binds DLL4 with a K_D of 10nM or less. In some embodiments, the DLL4-binding agent binds DLL4 with a K_D of about 1nM or less. In some embodiments, the DLL4-binding agent binds DLL4 with a K_D of about 0.1nM or less. In some embodiments, the DLL4-binding agent binds DLL4 with a K_D of about 0.01nM or less. In some embodiments, at least one amino acid residue in at least one CDR of the DLL4-binding agent is substituted with a different amino acid so that the affinity of the DLL4-binding agent for DLL4 is altered. In some embodiments, the affinity of the DLL4-binding agent is increased. In some embodiments, the affinity of the DLL4-binding agent is decreased.

[00233] In certain embodiments, the DLL4-binding agent comprises the heavy chain variable region and the light chain variable region of the 21R79 antibody. In certain embodiments, the DLL4-binding agent comprises the heavy chain and light chain of the 21R79 antibody (with or without the leader sequence). In certain embodiments, the DLL4-binding agent is the 21R79 antibody. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13236. The plasmid PTA-13232 was deposited with the ATCC, at 10801 University Boulevard, Manassas, VA, 20110, under the conditions of the Budapest Treaty on September 21, 2012. In some embodiments, the DLL4-binding agent comprises the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13232 and the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235.

[00234] In certain embodiments, a DLL4-binding agent comprises, consists essentially of, or consists of, the antibody 21R79.

[00235] In certain embodiments, the DLL4-binding agent comprises the heavy chain variable region and the light chain variable region of the 21R75 antibody. In certain embodiments, the DLL4-binding agent

comprises the heavy chain and light chain of the 21R75 antibody (with or without the leader sequence). In certain embodiments, the DLL4-binding agent is the 21R75 antibody. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13234. The plasmid PTA-13234 was deposited with the ATCC, at 10801 University Boulevard, Manassas, VA, 20110, under the conditions of the Budapest Treaty on September 21, 2012. In some embodiments, the DLL4-binding agent comprises the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13232 and the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235.

[00236] In certain embodiments, a DLL4-binding agent comprises, consists essentially of, or consists of, the antibody 21R75.

[00237] In certain embodiments, the DLL4-binding agent comprises the heavy chain variable region and the light chain variable region of the 21R83 antibody. In certain embodiments, the DLL4-binding agent comprises the heavy chain and light chain of the 21R83 antibody (with or without the leader sequence). In certain embodiments, the DLL4-binding agent is the 21R83 antibody. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13278. The plasmid PTA-13278 was deposited with the ATCC, at 10801 University Boulevard, Manassas, VA, 20110, under the conditions of the Budapest Treaty on October 24, 2012. In some embodiments, the DLL4-binding agent comprises the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235. In some embodiments, the DLL4-binding agent comprises the same heavy chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13278 and the same light chain variable region as a polypeptide encoded by the plasmid on deposit as ATCC Patent Deposit Designation PTA-13235.

[00238] In certain embodiments, a DLL4-binding agent comprises, consists essentially of, or consists of, the antibody 21R83.

[00239] In certain embodiments, a DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on DLL4 as an antibody of the invention. In another embodiment, a DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by an antibody of the invention. In certain embodiments, a DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R79. In another embodiment, the DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R79. In certain embodiments, a DLL4-binding agent (e.g., an antibody)

binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R75. In another embodiment, the DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R75. In certain embodiments, a DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R83. In another embodiment, the DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R83.

[00240] In some embodiments, the DLL4-binding agent inhibits binding of DLL4 to at least one Notch receptor. In certain embodiments, the Notch receptor is Notch1, Notch2, Notch3, or Notch4. In some embodiments, the DLL4-binding agent specifically binds DLL4 and inhibits DLL4 activity. In some embodiments, the DLL4-binding agent specifically binds DLL4 and inhibits Notch signaling. In some embodiments, the DLL4-binding agent specifically binds DLL4 and modulates angiogenesis. In some embodiments, the DLL4-binding agent specifically binds DLL4 and inhibits tumor growth. In some embodiments, the DLL4-binding agent specifically binds DLL4 and inhibits tumorigenicity. In some embodiments, the DLL4-binding agent specifically binds DLL4 and reduces the number or frequency of CSCs in a tumor.

[00241] In certain embodiments, the invention provides a VEGF/DLL4-binding agent that is a bispecific antibody. In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF. In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF and a second antigen-binding site that binds a tumor-associated target. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising: a first antigen-binding site that specifically binds human VEGF, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19). In some embodiments, the bispecific antibody further comprises: a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising: a first antigen-binding site that specifically binds human VEGF, wherein the first antigen-binding site comprises (a) a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00242] In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:11. In some

embodiments, the bispecific antibody further comprises a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:11, and a light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12.

[00243] In certain embodiments, the invention provides a VEGF/DLL4-binding agent that is a bispecific antibody. In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first antigen-binding site that specifically binds human DLL4. In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first antigen-binding site that specifically binds human DLL4 and a second antigen-binding site that binds a tumor-associated target. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody comprises a first antigen-binding site comprising a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody comprises a first antigen-binding site comprising a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISSYNGATNYNQKFKG (SEQ ID NO:15), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody comprises a first antigen-binding site comprising a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody comprises a first antigen-binding site comprising a heavy chain CDR1 comprising

TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16). In some embodiments, the bispecific antibody further comprises: a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody comprising: a first antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises (a) a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16), and (b) a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00244] In some embodiments, the VEGF/DLL4 binding agent is a bispecific antibody comprising a first heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64. In some embodiments, the bispecific antibody further comprises a light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64; and/or a light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12.

[00245] In certain embodiments, the invention provides a VEGF/DLL4-binding agent (e.g., a bispecific antibody) that specifically binds human VEGF and human DLL4. In some embodiments, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human VEGF, and b) a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19); wherein the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13) or AYYIH (SEQ ID NO:79), a heavy chain CDR2 comprising $YIX_1X_2YX_3X_4ATNYNQKFKG$ (SEQ ID NO:80), wherein X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid, and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, a

bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), YISSYNGATNYNQKFKG (SEQ ID NO:15), YIAGYKDATNYNQKFKG (SEQ ID NO:59), or YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

[00246] In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIANYNRATNYNQKFKG (SEQ ID NO:14), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody is 219R45-MB-21R79.

[00247] In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISSYNGATNYNQKFKG (SEQ ID NO:15), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody is 219R45-MB-21M18.

[00248] In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4,

wherein the first antigen-binding site which comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YIAGYKDATNYNQKFKG (SEQ ID NO:59), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody is 219R45-MB-21R75.

[00249] In some embodiments, the bispecific antibody comprises a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19), and the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and wherein both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22). In some embodiments, the bispecific antibody is 219R45-MB-21R83.

[00250] In some embodiments, the VEGF/DLL4 binding agent (e.g., a bispecific antibody) comprises a first heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 80% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64, and a first and a second light chain variable region having at least 80% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:11; a second heavy chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64; and a first and a second light chain variable region having at least about 85%, at least about 90%, at least about 95%, at least about 97%, or at least about 99% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:9, and a first and a second light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In

certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:10, and a first and a second light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:58, and a first and a second light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11, a second heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:64, and a first and a second light chain variable region having at least about 95% sequence identity to SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region comprising SEQ ID NO:11, a second heavy chain variable region comprising SEQ ID NO:9, and a first and a second light chain variable region comprising SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region comprising SEQ ID NO:11, a second heavy chain variable region comprising SEQ ID NO:10, and a first and a second light chain variable region comprising SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region comprising SEQ ID NO:11, a second heavy chain variable region comprising SEQ ID NO:58, and a first and a second light chain variable region comprising SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region comprising SEQ ID NO:11, a second heavy chain variable region comprising SEQ ID NO:64, and a first and a second light chain variable region comprising SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region consisting essentially of SEQ ID NO:11, a second heavy chain variable region consisting essentially of SEQ ID NO:9, and a first and a second light chain variable region consisting essentially of SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region consisting essentially of SEQ ID NO:11, a second heavy chain variable region consisting essentially of SEQ ID NO:10, and a first and a second light chain variable region consisting essentially of SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region consisting essentially of SEQ ID NO:11, a second heavy chain variable region consisting essentially of SEQ ID NO:58, and a first and a second light chain variable region consisting essentially of SEQ ID NO:12. In certain embodiments, the bispecific VEGF/DLL4-binding agent comprises a first heavy chain variable region consisting essentially of SEQ ID NO:11, a second heavy chain variable region

consisting essentially of SEQ ID NO:64, and a first and a second light chain variable region consisting essentially of SEQ ID NO:12.

[00251] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-VEGF antibody 219R45. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-DLL4 antibody 21M18. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-DLL4 antibody 21R79. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-DLL4 antibody 21R75. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-DLL4 antibody 21R83. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-VEGF antibody 219R45, a heavy chain variable region from the anti-DLL4 antibody 21R79 and two identical light chain variable regions. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-VEGF antibody 219R45, a heavy chain variable region from the anti-DLL4 antibody 21M18 and two identical light chain variable regions. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-VEGF antibody 219R45, a heavy chain variable region from the anti-DLL4 antibody 21R75 and two identical light chain variable regions. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain variable region from the anti-VEGF antibody 219R45, a heavy chain variable region from the anti-DLL4 antibody 21R83 and two identical light chain variable regions.

[00252] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first CH3 domain and a second CH3 domain, each of which is modified to promote formation of heteromultimers. In some embodiments, the first and second CH3 domains are modified using a knobs-into-holes technique. In some embodiments, the first and second CH3 domains comprise changes in amino acids that result in altered electrostatic interactions. In some embodiments, the first and second CH3 domains comprise changes in amino acids that result in altered hydrophobic/hydrophilic interactions.

[00253] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises heavy chain constant regions selected from the group consisting of: (a) a first human IgG1 constant region, wherein the amino acids at positions corresponding to positions 253 and 292 of SEQ ID NO:41 are replaced with glutamate or aspartate, and a second human IgG1 constant region, wherein the amino acids at positions corresponding to positions 240 and 282 of SEQ ID NO:41 are replaced with lysine; (b) a first human IgG2 constant region, wherein the amino acids at positions corresponding to positions 249 and 288 of SEQ I DNO42 are replaced with glutamate or aspartate, and a second human IgG2 constant region wherein the amino acids at positions corresponding to positions 236 and 278 of SEQ ID NO:42 are

replaced with lysine; (c) a first human IgG3 constant region, wherein the amino acids at positions corresponding to positions 300 and 339 of SEQ ID NO:43 are replaced with glutamate or aspartate, and a second human IgG3 constant region wherein the amino acids at positions corresponding to positions 287 and 329 of SEQ ID NO:43 are replaced with lysine; and (d) a first human IgG4 constant region, wherein the amino acids at positions corresponding to positions 250 and 289 of SEQ ID NO:44 are replaced with glutamate or aspartate, and a second IgG4 constant region wherein the amino acids at positions corresponding to positions 237 and 279 of SEQ ID NO:44 are replaced with lysine.

[00254] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG1 constant region with amino acid substitutions at positions corresponding to positions 253 and 292 of SEQ ID NO:41, wherein the amino acids are replaced with glutamate or aspartate, and a second human IgG1 constant region with amino acid substitutions at positions corresponding to positions 240 and 282 of SEQ ID NO:41, wherein the amino acids are replaced with lysine. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG2 constant region with amino acid substitutions at positions corresponding to positions 249 and 288 of SEQ ID NO:42, wherein the amino acids are replaced with glutamate or aspartate, and a second human IgG2 constant region with amino acid substitutions at positions corresponding to positions 236 and 278 of SEQ ID NO:42, wherein the amino acids are replaced with lysine. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG3 constant region with amino acid substitutions at positions corresponding to positions 300 and 339 of SEQ ID NO:43, wherein the amino acids are replaced with glutamate or aspartate, and a second human IgG2 constant region with amino acid substitutions at positions corresponding to positions 287 and 329 of SEQ ID NO:43, wherein the amino acids are replaced with lysine. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG4 constant region with amino acid substitutions at positions corresponding to positions 250 and 289 of SEQ ID NO:44, wherein the amino acids are replaced with glutamate or aspartate, and a second human IgG4 constant region with amino acid substitutions at positions corresponding to positions 237 and 279 of SEQ ID NO:44, wherein the amino acids are replaced with lysine.

[00255] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG2 constant region with amino acid substitutions at positions corresponding to positions 249 and 288 of SEQ ID NO:42, wherein the amino acids are replaced with glutamate, and a second human IgG2 constant region with amino acid substitutions at positions corresponding to positions 236 and 278 of SEQ ID NO:42, wherein the amino acids are replaced lysine. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a first human IgG2 constant region with amino acid substitutions at positions corresponding to positions 249 and 288 of SEQ ID NO:42, wherein the amino acids are replaced with aspartate, and a second human IgG2 constant region with amino acid

substitutions at positions corresponding to positions 236 and 278 of SEQ ID NO:42, wherein the amino acids are replaced with lysine.

[00256] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:7. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:5. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:56. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:62. In some embodiments, the bispecific antibody further comprises a light chain of SEQ ID NO:12. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:7, a heavy chain of SEQ ID NO:5, and two light chains of SEQ ID NO:8. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:7, a heavy chain of SEQ ID NO:6, and two light chains of SEQ ID NO:8. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:7, a heavy chain of SEQ ID NO:56, and two light chains of SEQ ID NO:8. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises a heavy chain of SEQ ID NO:7, a heavy chain of SEQ ID NO:62, and two light chains of SEQ ID NO:8.

[00257] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which binds VEGF with a K_D of about 50nM or less, about 25nM or less, about 10nM or less, about 1nM or less, or about 0.1nM or less. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which binds DLL4 with a K_D of about 50nM or less, about 25nM or less, about 10nM or less, about 1nM or less, or about 0.1nM or less. In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which binds VEGF with a K_D of about 50nM or less and binds DLL4 with a K_D of about 50nM or less. In some embodiments, the bispecific antibody binds VEGF with a K_D of about 25nM or less and binds DLL4 with a K_D of about 25nM or less. In some embodiments, the bispecific antibody binds VEGF with a K_D of about 10nM or less and binds DLL4 with a K_D of about 10nM or less. In some embodiments, the bispecific antibody binds VEGF with a K_D of about 1nM or less and binds DLL4 with a K_D of about 1nM or less.

[00258] In some embodiments, the VEGF/DLL4-binding agent is a bispecific antibody which comprises one antigen-binding site with a binding affinity that is weaker than the binding affinity of the second antigen-binding site. For example, in some embodiments, the bispecific antibody may bind VEGF with a K_D ranging from about 0.1nM to 1nM and may bind DLL4 with a K_D ranging from about 1nM to 10nM. Or the bispecific antibody may bind VEGF with a K_D ranging from about 1nM to 10nM and may bind DLL4 with a K_D ranging from about 0.1nM to 1nM. In some embodiments, the bispecific antibody may bind DLL4 with a K_D ranging from about 0.1nM to 1nM and may bind VEGF with a K_D ranging from about 1nM to 10nM. Or the bispecific antibody may bind DLL4 with a K_D ranging from about 1nM to

10nM and may bind VEGF with a K_D ranging from about 0.1nM to 1nM. In some embodiments, the difference in affinity between the two antigen-binding sites may be about 2-fold or more, about 3-fold or more, about 5-fold or more, about 8-fold or more, about 10-fold or more, about 15-fold or more, about 30-fold or more, about 50-fold or more, or about 100-fold or more. In some embodiments, at least one amino acid residue in at least one CDR of the antigen-binding site for VEGF is substituted with a different amino acid so that the affinity of the VEGF-binding site is altered. In some embodiments, the affinity of the VEGF-binding site is increased. In some embodiments, the affinity of the VEGF-binding site is decreased. In some embodiments, at least one amino acid residue in at least one CDR of the antigen-binding site for DLL4 is substituted with a different amino acid so that the affinity of the DLL4-binding site is altered. In some embodiments, the affinity of the DLL4-binding site is increased. In some embodiments, the affinity of the DLL4-binding site is decreased. In some embodiments, the affinities of both the VEGF and DLL4 antigen-binding sites are altered.

[00259] The invention provides polypeptides, including but not limited to antibodies, that specifically bind VEGF and/or DLL4. In some embodiments, a polypeptide binds human VEGF. In some embodiments, a polypeptide binds human DLL4. In some embodiments, a polypeptide binds human VEGF and mouse VEGF. In some embodiments, a polypeptide binds human DLL4 and mouse DLL4.

[00260] In some embodiments, a VEGF-binding agent comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:47, and SEQ ID NO:49.

[00261] In some embodiments, a DLL4-binding agent comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:4, SEQ ID NO:5, SEQ NO ID:6, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64.

[00262] In some embodiments, a VEGF/DLL4-binding agent comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64.

[00263] In some embodiments, a VEGF/DLL4-binding agent comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64. In some embodiments, the VEGF/DLL4 binding agent further comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:3, SEQ ID NO:7, SEQ ID NO:11, SEQ ID NO:47, and SEQ ID NO:49.

In some embodiments, the VEGF/DLL4 binding agent further comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:4, SEQ ID NO:8, and SEQ ID NO:12.

[00264] In some embodiments, a VEGF/DLL4-binding agent comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:3, SEQ ID NO:7, SEQ ID NO:11, SEQ ID NO:47, and SEQ ID NO:49. In some embodiments, the VEGF/DLL4 binding agent further comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:46, SEQ ID NO:48, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64. In some embodiments, the VEGF/DLL4 binding agent further comprises a polypeptide comprising a sequence selected from the group consisting of: SEQ ID NO:4, SEQ ID NO:8, and SEQ ID NO:12.

[00265] In certain embodiments, a VEGF/DLL4-binding agent (e.g., antibody) competes for specific binding to VEGF with an antibody that comprises a heavy chain variable region comprising SEQ ID NO:11 and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, a VEGF/DLL4-binding agent competes with antibody 219R45 for specific binding to human VEGF. In some embodiments, a VEGF/DLL4-binding agent or antibody competes for specific binding to VEGF in an *in vitro* competitive binding assay. In some embodiments, the VEGF is human VEGF. In some embodiments, the VEGF is mouse VEGF.

[00266] In certain embodiments, a VEGF-DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on VEGF as an antibody of the invention. In another embodiment, a VEGF/DLL4-binding agent is an antibody that binds an epitope on VEGF that overlaps with the epitope on VEGF bound by an antibody of the invention. In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on VEGF as antibody 219R45. In another embodiment, the VEGF/DLL4-binding agent is an antibody that binds an epitope on VEGF that overlaps with the epitope on VEGF bound by antibody 219R45.

[00267] In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to VEGF with the antibody 219R45 (e.g., in a competitive binding assay).

[00268] In certain embodiments, a VEGF/DLL4-binding agent (e.g., antibody) competes for specific binding to DLL4 with an antibody that comprises a heavy chain variable region comprising SEQ ID NO:9 SEQ ID NO:10, SEQ ID NO:58, or SEQ ID NO:64 and a light chain variable region comprising SEQ ID NO:12. In certain embodiments, a VEGF/DLL4-binding agent competes with antibody 21R79 for specific binding to human DLL4. In certain embodiments, a VEGF/DLL4-binding agent competes with antibody 21R75 for specific binding to human DLL4. In certain embodiments, a VEGF/DLL4-binding agent competes with antibody 21R83 for specific binding to human DLL4. In some embodiments, a VEGF/DLL4-binding agent or antibody competes for specific binding to DLL4 in an *in vitro* competitive

binding assay. In some embodiments, the DLL4 is human DLL4. In some embodiments, the DLL4 is mouse DLL4.

[00269] In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) binds the same epitope, or essentially the same epitope, on DLL4 as an antibody of the invention. In another embodiment, a VEGF/DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by an antibody of the invention. In certain embodiments, a VEGF/DLL4-binding agent binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R79. In certain embodiments, a VEGF/DLL4-binding agent binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R75. In certain embodiments, a VEGF/DLL4-binding agent binds the same epitope, or essentially the same epitope, on DLL4 as antibody 21R83. In another embodiment, the VEGF/DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R79. In another embodiment, the VEGF/DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R75. In another embodiment, the VEGF/DLL4-binding agent is an antibody that binds an epitope on DLL4 that overlaps with the epitope on DLL4 bound by antibody 21R83.

[00270] In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to DLL4 with the antibody 21R79 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to DLL4 with the antibody 21R75 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to DLL4 with the antibody 21R83 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to DLL4 with the antibody 21M18 (e.g., in a competitive binding assay).

[00271] In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to VEGF and/or DLL4 with the bispecific antibody 219R45-MB-21M18 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to VEGF and/or DLL4 with the bispecific antibody 219R45-MB-21M79 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to VEGF and/or DLL4 with the bispecific antibody 219R45-MB-21M75 (e.g., in a competitive binding assay). In certain embodiments, the VEGF/DLL4-binding agent is an agent that competes for specific binding to VEGF and/or DLL4 with the bispecific antibody 219R45-MB-21M83 (e.g., in a competitive binding assay).

[00272] In certain embodiments, the VEGF/DLL4-binding agent (e.g., an antibody) described herein binds VEGF and modulates VEGF activity. In some embodiments, the VEGF/DLL4-binding agent is a VEGF antagonist and inhibits VEGF activity. In some embodiments, the VEGF/DLL4-binding agent is a VEGF antagonist and modulates angiogenesis. In some embodiments, the VEGF/DLL4-binding agent is a

VEGF antagonist and inhibits angiogenesis. In some embodiments, the VEGF/DLL4-binding agent is a VEGF antagonist and inhibits tumor growth.

[00273] In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) described herein binds human DLL4 and modulates DLL4 activity. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and inhibits DLL4 activity. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and inhibits Notch activity. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and inhibits Notch signaling. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and modulates angiogenesis. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and promotes aberrant angiogenesis. In some embodiments, a VEGF/DLL4-binding agent is a DLL4 antagonist and inhibits tumor growth.

[00274] In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) described herein is a bispecific antibody that binds human VEGF and modulates VEGF activity. In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) described herein is a bispecific antibody that binds human DLL4 and modulates DLL4 activity. In certain embodiments, a VEGF/DLL4-binding agent (e.g., an antibody) described herein is a bispecific antibody that binds human VEGF and human DLL4 and modulates both VEGF and DLL4 activity. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and inhibits both VEGF activity and DLL4 activity. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and inhibits VEGF activity and Notch activity. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and inhibits VEGF activity and Notch signaling. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and modulates angiogenesis. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and promotes aberrant angiogenesis. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and inhibits angiogenesis. In some embodiments, the bispecific antibody is a VEGF antagonist and a DLL4 antagonist and inhibits tumor growth.

[00275] In certain embodiments, the VEGF/DLL4-binding agent (e.g., an antibody or a bispecific antibody) is an antagonist of VEGF. In some embodiments, the VEGF/DLL4-binding agent is an antagonist of VEGF and inhibits VEGF activity. In certain embodiments, the VEGF/DLL4-binding agent inhibits VEGF activity by at least about 10%, at least about 20%, at least about 30%, at least about 50%, at least about 75%, at least about 90%, or about 100%. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is antibody 219R45. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is a bispecific antibody comprising the antigen-binding site of 219R45. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is the bispecific antibody 219R45-MB-21M18. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is the bispecific antibody 219R45-MB-21R79. In certain

embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is the bispecific antibody 219R45-MB-21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human VEGF activity is the bispecific antibody 219R45-MB-21R83.

[00276] In certain embodiments, the VEGF/DLL4-binding agent (e.g., an antibody) is an antagonist of DLL4. In some embodiments, the VEGF/DLL4-binding agent is an antagonist of DLL4 and inhibits DLL4 activity. In certain embodiments, the VEGF/DLL4-binding agent inhibits DLL4 activity by at least about 10%, at least about 20%, at least about 30%, at least about 50%, at least about 75%, at least about 90%, or about 100%. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is antibody 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is antibody 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is antibody 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is a bispecific antibody comprising the antigen-binding site of 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is a bispecific antibody comprising the antigen-binding site of 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is a bispecific antibody comprising the antigen-binding site of 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is the bispecific antibody 219R45-MB-21M18. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is the bispecific antibody 219R45-MB-21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is the bispecific antibody 219R45-MB-21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits human DLL4 activity is the bispecific antibody 219R45-MB-21R83.

[00277] In certain embodiments, the VEGF/DLL4-binding agent (e.g., antibody) is an antagonist of Notch signaling. In certain embodiments, the VEGF/DLL4-binding agent inhibits Notch signaling by at least about 10%, at least about 20%, at least about 30%, at least about 50%, at least about 75%, at least about 90%, or about 100%. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is antibody 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is antibody 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is antibody 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is a bispecific antibody comprising the antigen-binding site of 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is a bispecific antibody comprising the antigen-binding site of 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is a bispecific antibody comprising the antigen-binding site of 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is the bispecific antibody 219R45-MB-21M18. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is the bispecific antibody 219R45-MB-21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch

signaling is the bispecific antibody 219R45-MB-21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits Notch signaling is the bispecific antibody 219R45-MB-21R83.

[00278] In certain embodiments, the VEGF/DLL4-binding agent (e.g., antibody) inhibits binding of VEGF to at least one receptor. In some embodiments, the VEGF/DLL4-binding agent inhibits binding of VEGF to VEGFR-1 or VEGFR-2. In certain embodiments, the VEGF/DLL4-binding agent inhibits binding of VEGF to at least one VEGF receptor by at least about 10%, at least about 25%, at least about 50%, at least about 75%, at least about 90%, or at least about 95%. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is antibody 219R45. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is a bispecific antibody comprising the antigen-binding site of 219R45. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is the bispecific antibody 219R45-MB-21M18. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is the bispecific antibody 219R45-MB-21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is the bispecific antibody 219R45-MB-21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human VEGF to at least one VEGF receptor is the bispecific antibody 219R45-MB-21R83.

[00279] In certain embodiments, the VEGF/DLL4-binding agent (e.g., antibody) inhibits binding of DLL4 protein to at least one Notch receptor. In some embodiments, the VEGF/DLL4-binding agent inhibits binding of DLL4 to Notch1, Notch2, Notch3, and/or Notch4. In certain embodiments, the VEGF/DLL4-binding agent inhibits binding of DLL4 to at least one Notch receptor by at least about 10%, at least about 25%, at least about 50%, at least about 75%, at least about 90%, or at least about 95%. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is antibody 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is antibody 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is antibody 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is a bispecific antibody comprising the antigen-binding site of 21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is a bispecific antibody comprising the antigen-binding site of 21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is a bispecific antibody comprising the antigen-binding site of 21R83. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is the bispecific antibody 219R45-MB-21M18. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is the bispecific antibody 219R45-MB-

21R79. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is the bispecific antibody 219R45-MB-21R75. In certain embodiments, a VEGF/DLL4-binding agent that inhibits binding of human DLL4 to at least one Notch receptor is the bispecific antibody 219R45-MB-21R83.

[00280] *In vivo* and *in vitro* assays for determining whether a VEGF/DLL4-binding agent (or candidate VEGF/DLL4-binding agent) inhibits VEGF or affects angiogenesis are known in the art. *In vitro* assays of angiogenesis include but are not limited to, HUVEC proliferation assays, endothelial cell tube formation assays, sprouting (or sprout formation) assays, HUVEC cell migration assays, and invasion assays. In some embodiments, cells in the presence of VEGF and the presence of a VEGF/DLL4-binding agent are compared to cells in the presence of VEGF without the VEGF/DLL4-binding agent present, and evaluated for effects on angiogenesis (or biological effects associated with angiogenesis). *In vivo* assays of angiogenesis include, but are not limited to, matrigel plug assays, corneal micropocket assays, and chicken chorioallantoic membrane (CAM) assays.

[00281] *In vivo* and *in vitro* assays for determining whether a VEGF/DLL4-binding agent (or candidate VEGF/DLL4-binding agent) inhibits Notch activation or signaling are known in the art. For example, cell-based, luciferase reporter assays utilizing a TCF/Luc reporter vector containing multiple copies of the TCF-binding domain upstream of a firefly luciferase reporter gene may be used to measure Notch signaling levels *in vitro* (Gazit et al., 1999, *Oncogene,* 18; 5959-66; TOPflash, Millipore, Billerica MA). In some embodiments, a cell-based, luciferase reporter assay utilizing a CBF/Luc reporter vector containing multiple copies of the CBF-binding domain upstream of a firefly luciferase report genes may be used. The level of Notch signaling in the presence of one or more Notch ligands (e.g., DLL4 expressed on the surface of transfected cells or soluble DLL4-Fc fusion protein) and in the presence of a VEGF/DLL4-binding agent is compared to the level of Notch signaling without the VEGF/DLL4-binding agent present.

[00282] In certain embodiments, the VEGF/DLL4-binding agents have one or more of the following effects: inhibit proliferation of tumor cells, inhibit tumor growth, reduce the tumorigenicity of a tumor, reduce the frequency of cancer stem cells in a tumor, trigger cell death of tumor cells, prevent metastasis of tumor cells, decrease survival of tumor cells, modulate angiogenesis, inhibit angiogenesis, inhibit productive angiogenesis, or promote aberrant angiogenesis.

[00283] In certain embodiments, the VEGF/DLL4-binding agents are capable of inhibiting tumor growth (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor). In certain embodiments, the VEGF/DLL4-binding agents are capable of inhibiting tumor growth *in vivo* (e.g., in a xenograft mouse model, and/or in a human having cancer). In certain embodiments, tumor growth is inhibited at least about two-fold, about three-fold, about five-fold, about ten-fold, about 50-fold, about 100-fold, or about 1000-fold as compared to an untreated tumor.

[00284] In certain embodiments, the VEGF/DLL4-binding agents are capable of reducing the tumorigenicity of a tumor (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor). In certain embodiments, the VEGF/DLL4-binding agent or antibody is capable of reducing the tumorigenicity of a tumor (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor) comprising cancer stem cells in an animal model, such as a mouse xenograft model. In certain embodiments, the VEGF/DLL4-binding agent or antibody is capable of reducing the tumorigenicity of a tumor (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor) by decreasing the number or frequency of cancer stem cells in the tumor. In certain embodiments, the number or frequency of cancer stem cells in a tumor (e.g., a colorectal, ovarian, pancreatic, or endometrial tumor) is reduced by at least about two-fold, about three-fold, about five-fold, about ten-fold, about 50-fold, about 100-fold, or about 1000-fold. In certain embodiments, the reduction in the number or frequency of cancer stem cells is determined by limiting dilution assay using an animal model. Additional examples and guidance regarding the use of limiting dilution assays to determine a reduction in the number or frequency of cancer stem cells in a tumor can be found, e.g., in International Publication Number WO 2008/042236; U.S. Patent Publication No. 2008/0064049; and U.S. Patent Publication No. 2008/0178305.

[00285] In certain embodiments, the VEGF/DLL4-binding agents are capable of modulating angiogenesis. In certain embodiments, the VEGF/DLL4-binding agents are capable of modulating angiogenesis *in vivo* (e.g., in a xenograft mouse model, and/or in a human having cancer). In certain embodiments, VEGF/DLL4-binding agents are capable of inhibiting angiogenesis. In certain embodiments, VEGF/DLL4-binding agents are capable of promoting aberrant angiogenesis. In certain embodiments, VEGF/DLL4-binding agents are capable of inhibiting angiogenesis and/or promoting aberrant angiogenesis, leading to unproductive vascularization.

[00286] In certain embodiments, the VEGF/DLL4-binding agents described herein have a circulating half-life in mice, cynomolgus monkeys, or humans of at least about 2 hours, at least about 5 hours, at least about 10 hours, at least about 24 hours, at least about 3 days, at least about 1 week, or at least about 2 weeks. In certain embodiments, the VEGF/DLL4-binding agent is an IgG (e.g., IgG1 or IgG2) antibody that has a circulating half-life in mice, cynomolgus monkeys, or humans of at least about 2 hours, at least about 5 hours, at least about 10 hours, at least about 24 hours, at least about 3 days, at least about 1 week, or at least about 2 weeks. Methods of increasing (or decreasing) the half-life of agents such as polypeptides and antibodies are known in the art. For example, known methods of increasing the circulating half-life of IgG antibodies include the introduction of mutations in the Fc region which increase the pH-dependent binding of the antibody to the neonatal Fc receptor (FcRn) at pH 6.0 (see, e.g., U.S. Patent Publication Nos. 2005/0276799, 2007/0148164, and 2007/0122403). Known methods of increasing the circulating half-life of antibody fragments lacking the Fc region include such techniques as PEGylation.

[00287] In some embodiments, the VEGF/DLL4-binding agents are antibodies. Polyclonal antibodies can be prepared by any known method. In some embodiments, polyclonal antibodies are produced by immunizing an animal (e.g., a rabbit, rat, mouse, goat, donkey) with an antigen of interest (e.g., a purified peptide fragment, full-length recombinant protein, or fusion protein) by multiple subcutaneous or intraperitoneal injections. The antigen can be optionally conjugated to a carrier such as keyhole limpet hemocyanin (KLH) or serum albumin. The antigen (with or without a carrier protein) is diluted in sterile saline and usually combined with an adjuvant (e.g., Complete or Incomplete Freund's Adjuvant) to form a stable emulsion. After a sufficient period of time, polyclonal antibodies are recovered from the immunized animal, usually from blood or ascites. The polyclonal antibodies can be purified from serum or ascites according to standard methods in the art including, but not limited to, affinity chromatography, ion-exchange chromatography, gel electrophoresis, and dialysis.

[00288] In some embodiments, the VEGF/DLL4-binding agents are monoclonal antibodies. Monoclonal antibodies can be prepared using hybridoma methods known to one of skill in the art. In some embodiments, using the hybridoma method, a mouse, hamster, or other appropriate host animal, is immunized as described above to elicit from lymphocytes the production of antibodies that specifically bind the immunizing antigen. In some embodiments, lymphocytes can be immunized *in vitro*. In some embodiments, the immunizing antigen can be a human protein or a portion thereof. In some embodiments, the immunizing antigen can be a mouse protein or a portion thereof.

[00289] Following immunization, lymphocytes are isolated and fused with a suitable myeloma cell line using, for example, polyethylene glycol. The hybridoma cells are selected using specialized media as known in the art and unfused lymphocytes and myeloma cells do not survive the selection process. Hybridomas that produce monoclonal antibodies directed specifically against a chosen antigen may be identified by a variety of methods including, but not limited to, immunoprecipitation, immunoblotting, and *in vitro* binding assays (e.g., flow cytometry, FACS, ELISA, and radioimmunoassay). The hybridomas can be propagated either in *in vitro* culture using standard tissue culture methods or *in vivo* as ascites tumors in an animal. The monoclonal antibodies can be purified from the culture medium or ascites fluid according to standard methods in the art including, but not limited to, affinity chromatography, ion-exchange chromatography, gel electrophoresis, and dialysis.

[00290] In certain embodiments, monoclonal antibodies can be made using recombinant DNA techniques as known to one skilled in the art. The polynucleotides encoding a monoclonal antibody are isolated from mature B-cells or hybridoma cells, such as by RT-PCR using oligonucleotide primers that specifically amplify the genes encoding the heavy and light chains of the antibody, and their sequence is determined using standard techniques. The isolated polynucleotides encoding the heavy and light chains are then cloned into suitable expression vectors which produce the monoclonal antibodies when transfected into

host cells such as E. coli, simian COS cells, Chinese hamster ovary (CHO) cells, or myeloma cells that do not otherwise produce immunoglobulin proteins.

[00291] In certain other embodiments, recombinant monoclonal antibodies, or fragments thereof, can be isolated from phage display libraries expressing variable domains or CDRs of a desired species.

[00292] The polynucleotide(s) encoding a monoclonal antibody can be modified, for example, by using recombinant DNA technology to generate alternative antibodies. In some embodiments, the constant domains of the light and heavy chains of, for example, a mouse monoclonal antibody can be substituted for those regions of, for example, a human antibody to generate a chimeric antibody, or for a non-immunoglobulin polypeptide to generate a fusion antibody. In some embodiments, the constant regions are truncated or removed to generate the desired antibody fragment of a monoclonal antibody. Site-directed or high-density mutagenesis of the variable region can be used to optimize specificity, affinity, etc. of a monoclonal antibody.

[00293] In some embodiments, a monoclonal antibody against VEGF and/or DLL4 is a humanized antibody. Typically, humanized antibodies are human immunoglobulins in which residues from the CDRs are replaced by residues from a CDR of a non-human species (e.g., mouse, rat, rabbit, hamster, etc.) that have the desired specificity, affinity, and/or binding capability using methods known to one skilled in the art. In some embodiments, the Fv framework region residues of a human immunoglobulin are replaced with the corresponding residues in an antibody from a non-human species that has the desired specificity, affinity, and/or binding capability. In some embodiments, a humanized antibody can be further modified by the substitution of additional residues either in the Fv framework region and/or within the replaced non-human residues to refine and optimize antibody specificity, affinity, and/or capability. In general, a humanized antibody will comprise substantially all of at least one, and typically two or three, variable domain regions containing all, or substantially all, of the CDRs that correspond to the non-human immunoglobulin whereas all, or substantially all, of the framework regions are those of a human immunoglobulin consensus sequence. In some embodiments, a humanized antibody can also comprise at least a portion of an immunoglobulin constant region or domain (Fc), typically that of a human immunoglobulin. In certain embodiments, such humanized antibodies are used therapeutically because they may reduce antigenicity and HAMA (human anti-mouse antibody) responses when administered to a human subject.

[00294] In certain embodiments, the VEGF/DLL4-binding agent is a human antibody. Human antibodies can be directly prepared using various techniques known in the art. In some embodiments, human antibodies may be generated from immortalized human B lymphocytes immunized *in vitro* or from lymphocytes isolated from an immunized individual. In either case, cells that produce an antibody directed against a target antigen can be generated and isolated by techniques known in the art. In some embodiments, a human antibody can be selected from a phage library, where that phage library expresses

human antibodies. Alternatively, phage display technology can be used to produce human antibodies and antibody fragments *in vitro*, from immunoglobulin variable domain gene repertoires from unimmunized donors. Techniques for the generation and use of antibody phage libraries are well-known by those of skill in the art. Once antibodies are identified, affinity maturation strategies known in the art, including but not limited to, chain shuffling and site-directed mutagenesis, may be employed to generate high affinity human antibodies.

[00295] In some embodiments, human antibodies can be made in transgenic mice that contain human immunoglobulin loci. Upon immunization these mice are capable of producing the full repertoire of human antibodies in the absence of endogenous immunoglobulin production.

[00296] This invention also encompasses bispecific antibodies. Bispecific antibodies are capable of specifically recognizing and binding at least two different antigens or epitopes. The different epitopes can either be within the same molecule (e.g., two epitopes on a single protein) or on different molecules (e.g., one epitope on a protein and one epitope on a second protein). In some embodiments, a bispecific antibody has enhanced potency as compared to an individual antibody or to a combination of more than one antibody. In some embodiments, a bispecific antibody has reduced toxicity as compared to an individual antibody or to a combination of more than one antibody. It is known to those of skill in the art that any binding agent (e.g., antibody) may have unique pharmacokinetics (PK) (e.g., circulating half-life). In some embodiments, a bispecific antibody has the ability to synchronize the PK of two active binding agents wherein the two individual binding agents have different PK profiles. In some embodiments, a bispecific antibody has the ability to concentrate the actions of two binding agents (e.g., antibodies) in a common area (e.g., a tumor and/or tumor environment). In some embodiments, a bispecific antibody has the ability to concentrate the actions of two binding agents (e.g., antibodies) to a common target (e.g., a tumor or a tumor cell). In some embodiments, a bispecific antibody has the ability to target the actions of two binding agents (e.g., antibodies) to more than one biological pathway or function.

[00297] In certain embodiments, the bispecific antibody specifically binds VEGF and a second target. In certain embodiments, the bispecific antibody specifically binds DLL4 and a second target. In certain embodiments, the bispecific antibody specifically binds VEGF and DLL4. In some embodiments, the bispecific antibody specifically binds human VEGF and human DLL4. In some embodiments, the bispecific antibody is a monoclonal human or a humanized antibody. In some embodiments, the bispecific antibody inhibits angiogenesis and reduces cancer stem cell number or frequency. In some embodiments, the bispecific antibody inhibits blood vessel growth and inhibits blood vessel maturation. In some embodiments, the bispecific antibody prevents endothelial hyperproliferation. In some embodiments, the bispecific antibody has decreased toxicity and/or side effects. In some embodiments, the bispecific antibody has decreased toxicity and/or side effects as compared to a mixture of the two

individual antibodies or the antibodies as single agents. In some embodiments, the bispecific antibody has an increased therapeutic index. In some embodiments, the bispecific antibody has an increased therapeutic index as compared to a mixture of the two individual antibodies or the antibodies as single agents.

[00298] In some embodiments, the bispecific antibody can specifically recognize and bind a first antigen target, (e.g., DLL4) as well as a second antigen target, such as an effector molecule on a leukocyte (e.g., CD2, CD3, CD28, CD80, or CD87) or a Fc receptor (e.g., CD64, CD32, or CD16) so as to focus cellular defense mechanisms to the cell expressing the first antigen target. In some embodiments, the bispecific antibodies can be used to direct cytotoxic agents to cells which express a particular target antigen. These antibodies possess an antigen-binding site (e.g., to human DLL4) and a second site which binds a cytotoxic agent or a radionuclide chelator, such as EOTUBE, DPTA, DOTA, or TETA.

[00299] Techniques for making bispecific antibodies are known by those skilled in the art, see for example, Millstein et al., 1983, *Nature*, 305:537-539; Brennan et al., 1985, *Science*, 229:81; Suresh et al., 1986, *Methods in Enzymol.*, 121:120; Traunecker et al., 1991, *EMBO J.,* 10:3655-3659; Shalaby et al., 1992, *J. Exp. Med.,* 175:217-225; Kostelny et al., 1992, *J. Immunol.,* 148:1547-1553; Gruber et al., 1994, *J. Immunol.,* 152:5368; U.S. Patent No. 5,731,168; International Publication No. WO 2009/089004; and U.S. Patent Publication No. 2011/0123532. In some embodiments, the bispecific antibodies comprise heavy chain constant regions with modifications in the amino acids which are part of the interface between the two heavy chains. In some embodiments, the bispecific antibodies can be generated using a "knobs-into-holes" strategy (see. e.g., U.S. Patent No. 5,731,168; Ridgway et. al., 1996, *Prot. Engin.*, 9:617-621). At times, the "knobs" and "holes" terminology is replaced with the terms "protuberances" and "cavities". In some embodiments, the bispecific antibodies may comprise variant hinge regions incapable of forming disulfide linkages between the heavy chains (see, e.g., WO 2006/028936). In some embodiments, the modifications may comprise changes in amino acids that result in altered electrostatic interactions. In some embodiments, the modifications may comprise changes in amino acids that result in altered hydrophobic/hydrophilic interactions.

[00300] Bispecific antibodies can be intact antibodies or antibody fragments comprising antigen-binding sites. Antibodies with more than two valencies are also contemplated. For example, trispecific antibodies can be prepared (Tutt et al., 1991, *J. Immunol.*, 147:60). Thus, in certain embodiments the antibodies to VEGF and/or DLL4 are multispecific.

[00301] In certain embodiments, the antibodies (or other polypeptides) described herein may be monospecific. In certain embodiments, each of the one or more antigen-binding sites that an antibody contains is capable of binding (or binds) a homologous epitope on different proteins.

[00302] In certain embodiments, the VEGF/DLL4-binding agent is an antibody fragment. Antibody fragments may have different functions or capabilities than intact antibodies; for example, antibody

fragments can have increased tumor penetration. Various techniques are known for the production of antibody fragments including, but not limited to, proteolytic digestion of intact antibodies. In some embodiments, antibody fragments include a $F(ab')_2$ fragment produced by pepsin digestion of an antibody molecule. In some embodiments, antibody fragments include a Fab fragment generated by reducing the disulfide bridges of an $F(ab')_2$ fragment. In other embodiments, antibody fragments include a Fab fragment generated by the treatment of the antibody molecule with papain and a reducing agent. In certain embodiments, antibody fragments are produced recombinantly. In some embodiments, antibody fragments include Fv or single chain Fv (scFv) fragments. Fab, Fv, and scFv antibody fragments can be expressed in and secreted from E. coli or other host cells, allowing for the production of large amounts of these fragments. In some embodiments, antibody fragments are isolated from antibody phage libraries as discussed herein. For example, methods can be used for the construction of Fab expression libraries (Huse et al., 1989, *Science*, 246:1275-1281) to allow rapid and effective identification of monoclonal Fab fragments with the desired specificity for VEGF and/or DLL4 or derivatives, fragments, analogs or homologs thereof. In some embodiments, antibody fragments are linear antibody fragments. In certain embodiments, antibody fragments are monospecific or bispecific. In certain embodiments, the VEGF/DLL4-binding agent is a scFv. Various techniques known to those of skill in the art can be used for the production of single-chain antibodies specific to VEGF or DLL4.

[00303] It can further be desirable, especially in the case of antibody fragments, to modify an antibody in order to alter (e.g., increase or decrease) its serum half-life. This can be achieved, for example, by incorporation of a salvage receptor binding epitope into the antibody fragment by mutation of the appropriate region in the antibody fragment or by incorporating the epitope into a peptide tag that is then fused to the antibody fragment at either end or in the middle (e.g., by DNA or peptide synthesis).

[00304] Heteroconjugate antibodies are also within the scope of the present invention. Heteroconjugate antibodies are composed of two covalently joined antibodies. Such antibodies have, for example, been proposed to target immune cells to unwanted cells (see, e.g., U.S. Patent No. 4,676,980). It is also contemplated that the heteroconjugate antibodies can be prepared *in vitro* using known methods in synthetic protein chemistry, including those involving cross-linking agents. For example, immunotoxins can be constructed using a disulfide exchange reaction or by forming a thioether bond. Examples of suitable reagents for this purpose include iminothiolate and methyl-4-mercaptobutyrimidate.

[00305] For the purposes of the present invention, it should be appreciated that modified antibodies can comprise any type of variable region that provides for the association of the antibody with the target (i.e., human VEGF or human DLL4). In this regard, the variable region may comprise or be derived from any type of mammal that can be induced to mount a humoral response and generate immunoglobulins against the desired antigen. As such, the variable region of the modified antibodies can be, for example, of human, murine, non-human primate (e.g. cynomolgus monkeys, macaques, etc.) or rabbit origin. In some

embodiments, both the variable and constant regions of the modified immunoglobulins are human. In other embodiments, the variable regions of compatible antibodies (usually derived from a non-human source) can be engineered or specifically tailored to improve the binding properties or reduce the immunogenicity of the molecule. In this respect, variable regions useful in the present invention can be humanized or otherwise altered through the inclusion of imported amino acid sequences.

[00306] In certain embodiments, the variable domains in both the heavy and light chains are altered by at least partial replacement of one or more CDRs and, if necessary, by partial framework region replacement and sequence modification and/or alteration. Although the CDRs may be derived from an antibody of the same class or even subclass as the antibody from which the framework regions are derived, it is envisaged that the CDRs may be derived from an antibody of different class and often from an antibody from a different species. It may not be necessary to replace all of the CDRs with all of the CDRs from the donor variable region to transfer the antigen binding capacity of one variable domain to another. Rather, it may only be necessary to transfer those residues that are required to maintain the activity of the antigen-binding site.

[00307] Alterations to the variable region notwithstanding, those skilled in the art will appreciate that the modified antibodies of this invention will comprise antibodies (e.g., full-length antibodies or immunoreactive fragments thereof) in which at least a fraction of one or more of the constant region domains has been deleted or otherwise altered so as to provide desired biochemical characteristics such as increased tumor localization or increased serum half-life when compared with an antibody of approximately the same immunogenicity comprising a native or unaltered constant region. In some embodiments, the constant region of the modified antibodies will comprise a human constant region. Modifications to the constant region compatible with this invention comprise additions, deletions or substitutions of one or more amino acids in one or more domains. The modified antibodies disclosed herein may comprise alterations or modifications to one or more of the three heavy chain constant domains (CH1, CH2 or CH3) and/or to the light chain constant domain (CL). In some embodiments, one or more domains are partially or entirely deleted from the constant regions of the modified antibodies. In some embodiments, the modified antibodies will comprise domain deleted constructs or variants wherein the entire CH2 domain has been removed (ΔCH2 constructs). In some embodiments, the omitted constant region domain is replaced by a short amino acid spacer (e.g., 10 amino acid residues) that provides some of the molecular flexibility typically imparted by the absent constant region.

[00308] In some embodiments, the modified antibodies are engineered to fuse the CH3 domain directly to the hinge region of the antibody. In other embodiments, a peptide spacer is inserted between the hinge region and the modified CH2 and/or CH3 domains. For example, constructs may be expressed wherein the CH2 domain has been deleted and the remaining CH3 domain (modified or unmodified) is joined to the hinge region with a 5-20 amino acid spacer. Such a spacer may be added to ensure that the regulatory

elements of the constant domain remain free and accessible or that the hinge region remains flexible. However, it should be noted that amino acid spacers may, in some cases, prove to be immunogenic and elicit an unwanted immune response against the construct. Accordingly, in certain embodiments, any spacer added to the construct will be relatively non-immunogenic so as to maintain the desired biological qualities of the modified antibodies.

[00309] In some embodiments, the modified antibodies may have only a partial deletion of a constant domain or substitution of a few or even a single amino acid. For example, the mutation of a single amino acid in selected areas of the CH2 domain may be enough to substantially reduce Fc binding and thereby increase cancer cell localization and/or tumor penetration. Similarly, it may be desirable to simply delete the part of one or more constant region domains that control a specific effector function (e.g. complement C1q binding) to be modulated. Such partial deletions of the constant regions may improve selected characteristics of the antibody (serum half-life) while leaving other desirable functions associated with the subject constant region domain intact. Moreover, as alluded to above, the constant regions of the disclosed antibodies may be modified through the mutation or substitution of one or more amino acids that enhances the profile of the resulting construct. In this respect it may be possible to disrupt the activity provided by a conserved binding site (e.g., Fc binding) while substantially maintaining the configuration and immunogenic profile of the modified antibody. In certain embodiments, the modified antibodies comprise the addition of one or more amino acids to the constant region to enhance desirable characteristics such as decreasing or increasing effector function or provide for more cytotoxin or carbohydrate attachment sites.

[00310] It is known in the art that the constant region mediates several effector functions. For example, binding of the C1 component of complement to the Fc region of IgG or IgM antibodies (bound to antigen) activates the complement system. Activation of complement is important in the opsonization and lysis of cell pathogens. The activation of complement also stimulates the inflammatory response and can also be involved in autoimmune hypersensitivity. In addition, the Fc region of an antibody can bind a cell expressing a Fc receptor (FcR). There are a number of Fc receptors which are specific for different classes of antibody, including IgG (gamma receptors), IgE (epsilon receptors), IgA (alpha receptors) and IgM (mu receptors). Binding of antibody to Fc receptors on cell surfaces triggers a number of important and diverse biological responses including engulfment and destruction of antibody-coated particles, clearance of immune complexes, lysis of antibody-coated target cells by killer cells (called antibody-dependent cell cytotoxicity or ADCC), release of inflammatory mediators, placental transfer, and control of immunoglobulin production.

[00311] In certain embodiments, the modified antibodies provide for altered effector functions that, in turn, affect the biological profile of the administered antibody. For example, in some embodiments, the deletion or inactivation (through point mutations or other means) of a constant region domain may reduce

Fc receptor binding of the circulating modified antibody thereby increasing cancer cell localization and/or tumor penetration. In other embodiments, the constant region modifications increase the serum half-life of the antibody. In other embodiments, the constant region modifications reduce the serum half-life of the antibody. In some embodiments, the constant region is modified to eliminate disulfide linkages or oligosaccharide moieties. Modifications to the constant region in accordance with this invention may easily be made using well known biochemical or molecular engineering techniques known to those of skill in the art.

[00312] In certain embodiments, a VEGF/DLL4-binding agent that is an antibody does not have one or more effector functions. For instance, in some embodiments, the antibody has no ADCC activity, and/or no complement-dependent cytotoxicity (CDC) activity. In certain embodiments, the antibody does not bind an Fc receptor, and/or complement factors. In certain embodiments, the antibody has no effector function.

[00313] The present invention further embraces variants and equivalents which are substantially homologous to the chimeric, humanized, and human antibodies, or antibody fragments thereof, set forth herein. These can contain, for example, conservative substitution mutations, i.e. the substitution of one or more amino acids by similar amino acids. For example, conservative substitution refers to the substitution of an amino acid with another amino acid within the same general class such as, for example, one acidic amino acid with another acidic amino acid, one basic amino acid with another basic amino acid or one neutral amino acid by another neutral amino acid. What is intended by a conservative amino acid substitution is well known in the art and described herein.

[00314] Thus, the present invention provides methods for producing an antibody that binds VEGF and/or DLL4, including bispecific antibodies that specifically bind both VEGF and DLL4. In some embodiments, the method for producing an antibody that binds VEGF and/or DLL4 comprises using hybridoma techniques. In some embodiments, the method of generating an antibody that binds VEGF or DLL4 or a bispecific antibody that binds VEGF and DLL4 comprises screening a human phage library. The present invention further provides methods of identifying an antibody that binds VEGF and/or DLL4. In some embodiments, the antibody is identified by FACS screening for binding to VEGF or a portion thereof. In some embodiments, the antibody is identified by FACS screening for binding to DLL4 or a portion thereof. In some embodiments, the antibody is identified by FACS screening for binding to both VEGF and DLL4 or a portion thereof. In some embodiments, the antibody is identified by screening using ELISA for binding to VEGF. In some embodiments, the antibody is identified by screening using ELISA for binding to DLL4. In some embodiments, the antibody is identified by screening using ELISA for binding to VEGF and DLL4. In some embodiments, the antibody is identified by FACS screening for blocking of binding of human VEGF to a human VEGF receptor. In some embodiments, the antibody is identified by FACS screening for blocking of binding of human DLL4 to a human Notch receptor. In

some embodiments, the antibody is identified by screening for inhibition or blocking of Notch signaling. In some embodiments, the antibody is identified by screening for inhibition or blocking of VEGF activity (e.g., induction of HUVEC proliferation). In some embodiments, the antibody is identified by screening for modulation of angiogenesis.

[00315] In certain embodiments, the antibodies described herein are isolated. In certain embodiments, the antibodies described herein are substantially pure.

[00316] In some embodiments of the present invention, the VEGF/DLL4-binding agents are polypeptides. The polypeptides can be recombinant polypeptides, natural polypeptides, or synthetic polypeptides comprising an antibody, or fragment thereof, that bind VEGF and/or DLL4. It will be recognized in the art that some amino acid sequences of the binding agents described herein can be varied without significant effect on the structure or function of the protein. Thus, the invention further includes variations of the polypeptides which show substantial activity or which include regions of an antibody, or fragment thereof, against human VEGF and/or DLL4. In some embodiments, amino acid sequence variations of VEGF/DLL4-binding polypeptides include deletions, insertions, inversions, repeats, and/or other types of substitutions.

[00317] In some embodiments, the polypeptides described herein are isolated. In some embodiments, the polypeptides described herein are substantially pure.

[00318] The polypeptides, analogs and variants thereof, can be further modified to contain additional chemical moieties not normally part of the polypeptide. The derivatized moieties can improve or otherwise modulate the solubility, the biological half-life, and/or absorption of the polypeptide. The moieties can also reduce or eliminate undesirable side effects of the polypeptides and variants. An overview for chemical moieties can be found in *Remington: The Science and Practice of Pharmacy, 22st Edition,* 2012, Pharmaceutical Press, London.

[00319] The polypeptides described herein can be produced by any suitable method known in the art. Such methods range from direct protein synthesis methods to constructing a DNA sequence encoding polypeptide sequences and expressing those sequences in a suitable host. In some embodiments, a DNA sequence is constructed using recombinant technology by isolating or synthesizing a DNA sequence encoding a wild-type protein of interest. Optionally, the sequence can be mutagenized by site-specific mutagenesis to provide functional analogs thereof.

[00320] In some embodiments, a DNA sequence encoding a polypeptide of interest may be constructed by chemical synthesis using an oligonucleotide synthesizer. Oligonucleotides can be designed based on the amino acid sequence of the desired polypeptide and selecting those codons that are favored in the host cell in which the recombinant polypeptide of interest will be produced. Standard methods can be applied to synthesize a polynucleotide sequence encoding an isolated polypeptide of interest. For example, a complete amino acid sequence can be used to construct a back-translated gene. Further, a DNA oligomer

containing a nucleotide sequence coding for the particular isolated polypeptide can be synthesized. For example, several small oligonucleotides coding for portions of the desired polypeptide can be synthesized and then ligated. The individual oligonucleotides typically contain 5' or 3' overhangs for complementary assembly.

[00321] Once assembled (by synthesis, site-directed mutagenesis, or another method), the polynucleotide sequences encoding a particular polypeptide of interest can be inserted into an expression vector and operatively linked to an expression control sequence appropriate for expression of the protein in a desired host. Proper assembly can be confirmed by nucleotide sequencing, restriction enzyme mapping, and/or expression of a biologically active polypeptide in a suitable host. As is well-known in the art, in order to obtain high expression levels of a transfected gene in a host, the gene must be operatively linked to transcriptional and translational expression control sequences that are functional in the chosen expression host.

[00322] In certain embodiments, recombinant expression vectors are used to amplify and express DNA encoding antibodies, or fragments thereof, against human VEGF and/or DLL4. For example, recombinant expression vectors can be replicable DNA constructs which have synthetic or cDNA-derived DNA fragments encoding a polypeptide chain of a VEGF/DLL4-binding agent, such as an anti-VEGF antibody or an anti-DLL4 antibody, or fragment thereof, operatively linked to suitable transcriptional and/or translational regulatory elements derived from mammalian, microbial, viral, or insect genes. A transcriptional unit generally comprises an assembly of (1) a genetic element or elements having a regulatory role in gene expression, for example, transcriptional promoters or enhancers, (2) a structural or coding sequence which is transcribed into mRNA and translated into protein, and (3) appropriate transcription and translation initiation and termination sequences. Regulatory elements can include an operator sequence to control transcription. The ability to replicate in a host, usually conferred by an origin of replication, and a selection gene to facilitate recognition of transformants can additionally be incorporated. DNA regions are "operatively linked" when they are functionally related to each other. For example, DNA for a signal peptide (secretory leader) is operatively linked to DNA for a polypeptide if it is expressed as a precursor which participates in the secretion of the polypeptide; a promoter is operatively linked to a coding sequence if it controls the transcription of the sequence; or a ribosome binding site is operatively linked to a coding sequence if it is positioned so as to permit translation. In some embodiments, structural elements intended for use in yeast expression systems include a leader sequence enabling extracellular secretion of translated protein by a host cell. In other embodiments, in situations where recombinant protein is expressed without a leader or transport sequence, it can include an N-terminal methionine residue. This residue can optionally be subsequently cleaved from the expressed recombinant protein to provide a final product.

[00323] The choice of an expression control sequence and an expression vector depends upon the choice of host. A wide variety of expression host/vector combinations can be employed. Useful expression vectors for eukaryotic hosts include, for example, vectors comprising expression control sequences from SV40, bovine papilloma virus, adenovirus, and cytomegalovirus. Useful expression vectors for bacterial hosts include known bacterial plasmids, such as plasmids from E. coli, including pCR1, pBR322, pMB9, and their derivatives, and wider host range plasmids, such as M13 and other filamentous single-stranded DNA phages.

[00324] The VEGF/DLL4-binding agents (e.g., polypeptides) of the present invention can be expressed from one or more vectors. For example, in some embodiments, one heavy chain polypeptide is expressed by one vector, a second heavy chain polypeptide is expressed by a second vector and a light chain polypeptide is expressed by a third vector. In some embodiments, a first heavy chain polypeptide and a light chain polypeptide is expressed by one vector and a second heavy chain polypeptide is expressed by a second vector. In some embodiments, two heavy chain polypeptides are expressed by one vector and a light chain polypeptide is expressed by a second vector. In some embodiments, three polypeptides are expressed from one vector. Thus, in some embodiments, a first heavy chain polypeptide, a second heavy chain polypeptide, and a light chain polypeptide are expressed by a single vector.

[00325] Suitable host cells for expression of a VEGF/DLL4-binding polypeptide or antibody (or a VEGF or DLL4 protein to use as an antigen) include prokaryotes, yeast cells, insect cells, or higher eukaryotic cells under the control of appropriate promoters. Prokaryotes include gram-negative or gram-positive organisms, for example E. coli or Bacillus. Higher eukaryotic cells include established cell lines of mammalian origin as described below. Cell-free translation systems may also be employed. Appropriate cloning and expression vectors for use with bacterial, fungal, yeast, and mammalian cellular hosts are described in Pouwels et al., 1985, *Cloning Vectors: A Laboratory Manual*, Elsevier, New York, NY. Additional information regarding methods of protein production, including antibody production, can be found, e.g., in U.S. Patent Publication No. 2008/0187954; U.S. Patent Nos. 6,413,746; 6,660,501; and International Patent Publication No. WO 04/009823.

[00326] Various mammalian or insect cell culture systems may be used to express recombinant polypeptides. Expression of recombinant proteins in mammalian cells may be desirable because these proteins are generally correctly folded, appropriately modified, and biologically functional. Examples of suitable mammalian host cell lines include, but are not limited to, COS-7 (monkey kidney-derived), L-929 (murine fibroblast-derived), C127 (murine mammary tumor-derived), 3T3 (murine fibroblast-derived), CHO (Chinese hamster ovary-derived), HeLa (human cervical cancer-derived), BHK (hamster kidney fibroblast-derived), HEK-293 (human embryonic kidney-derived) cell lines and variants of these cell lines. Mammalian expression vectors can comprise non-transcribed elements such as an origin of replication, a suitable promoter and enhancer linked to the gene to be expressed, and other 5' or 3' flanking

non-transcribed sequences, and 5' or 3' non-translated sequences, such as necessary ribosome binding sites, a polyadenylation site, splice donor and acceptor sites, and transcriptional termination sequences.

[00327] Expression of recombinant proteins in insect cell culture systems (e.g., baculovirus) also offers a robust method for producing correctly folded and biologically functional proteins. Baculovirus systems for production of heterologous proteins in insect cells are well-known to those of skill in the art.

[00328] Thus, the present invention provides cells comprising the VEGF/DLL4-binding agents described herein. In some embodiments, the cells produce the VEGF/DLL4-binding agents described herein. In certain embodiments, the cells produce an antibody. In some embodiments, the cells produce a VEGF-binding agent, such as an anti-VEGF antibody. In some embodiments, the cells produce a bispecific antibody that binds VEGF. In some embodiments, the cells produce a DLL4-binding agent, such as an anti-DLL4 antibody. In some embodiments, the cells produce a bispecific antibody that binds DLL4. In certain embodiments, the cells produce a bispecific VEGF/DLL4-binding agent, such as a bispecific antibody that binds VEGF and DLL4. In certain embodiments, the cells produce antibody 219R45. In certain embodiments, the cells produce antibody 21R79. In certain embodiments, the cells produce antibody 21R75. In certain embodiments, the cells produce antibody 21R83. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 219R45. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 21R79. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 21R75. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 21R83. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 219R45 and an antigen-binding site from antibody 21R79. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 219R45 and an antigen-binding site from antibody 21M18. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 219R45 and an antigen-binding site from antibody 21R75. In certain embodiments, the cells produce a bispecific antibody which comprises an antigen-binding site from antibody 219R45 and an antigen-binding site from antibody 21R83. In certain embodiments, the cells produce the bispecific antibody 219R45-MB-21M18. In certain embodiments, the cells produce the bispecific antibody 219R45-MB-21R79. In certain embodiments, the cells produce the bispecific antibody 219R45-MB-21R75. In certain embodiments, the cells produce the bispecific antibody 219R45-MB-21R83.

[00329] The proteins produced by a transformed host can be purified according to any suitable method. Standard methods include chromatography (e.g., ion exchange, affinity, and sizing column chromatography), centrifugation, differential solubility, or by any other standard technique for protein purification. Affinity tags such as hexa-histidine, maltose binding domain, influenza coat sequence, and

glutathione-S-transferase can be attached to the protein to allow easy purification by passage over an appropriate affinity column. Affinity chromatography used for purifying immunoglobulins can include Protein A, Protein G, and Protein L chromatography. Isolated proteins can be physically characterized using such techniques as proteolysis, size exclusion chromatography (SEC), mass spectrometry (MS), nuclear magnetic resonance (NMR), isoelectric focusing (IEF), high performance liquid chromatography (HPLC), and x-ray crystallography. The purity of isolated proteins can be determined using techniques known to those of skill in the art, including but not limited to, SDS-PAGE, SEC, capillary gel electrophoresis, IEF, and capillary isoelectric focusing (cIEF).

[00330] In some embodiments, supernatants from expression systems which secrete recombinant protein into culture media can be first concentrated using a commercially available protein concentration filter, for example, an Amicon or Millipore Pellicon ultrafiltration unit. Following the concentration step, the concentrate can be applied to a suitable purification matrix. In some embodiments, an anion exchange resin can be employed, for example, a matrix or substrate having pendant diethylaminoethyl (DEAE) groups. The matrices can be acrylamide, agarose, dextran, cellulose, or other types commonly employed in protein purification. In some embodiments, a cation exchange step can be employed. Suitable cation exchangers include various insoluble matrices comprising sulfopropyl or carboxymethyl groups. In some embodiments, a hydroxyapatite media can be employed, including but not limited to, ceramic hydroxyapatite (CHT). In certain embodiments, one or more reverse-phase HPLC steps employing hydrophobic RP-HPLC media, e.g., silica gel having pendant methyl or other aliphatic groups, can be employed to further purify a recombinant protein (e.g., a VEGF/DLL4-binding agent). Some or all of the foregoing purification steps, in various combinations, can be employed to provide a homogeneous recombinant protein.

[00331] In some embodiments, heterodimeric proteins such as bispecific antibodies are purified according the any of the methods described herein. In some embodiments, anti-VEGF/anti-DLL4 bispecific antibodies are isolated and/or purified using at least one chromatography step. In some embodiments, the at least one chromatography step comprises affinity chromatography. In some embodiments, the at least one chromatography step further comprises anion exchange chromatography. In some embodiments, the isolated and/or purified antibody product comprises at least 90% heterodimeric antibody. In some embodiments, the isolated and/or purified antibody product comprises at least 95%, 96%, 97%, 98% or 99% heterodimeric antibody. In some embodiments, the isolated and/or purified antibody product comprises about 100% heterodimeric antibody.

[00332] In some embodiments, recombinant protein produced in bacterial culture can be isolated, for example, by initial extraction from cell pellets, followed by one or more concentration, salting-out, aqueous ion exchange, or size exclusion chromatography steps. HPLC can be employed for final purification steps. Microbial cells employed in expression of a recombinant protein can be disrupted by

any convenient method, including freeze-thaw cycling, sonication, mechanical disruption, or use of cell lysing agents.

[00333] Methods known in the art for purifying antibodies and other proteins also include, for example, those described in U.S. Patent Publication Nos. 2008/0312425; 2008/0177048; and 2009/0187005.

[00334] In certain embodiments, the VEGF/DLL4-binding agent is a polypeptide that is not an antibody. A variety of methods for identifying and producing non-antibody polypeptides that bind with high affinity to a protein target are known in the art. See, e.g., Skerra, 2007, *Curr. Opin. Biotechnol.,* 18:295-304; Hosse et al., 2006, *Protein Science*, 15:14-27; Gill et al., 2006, *Curr. Opin. Biotechnol.*, 17:653-658; Nygren, 2008, *FEBSJ.,* 275:2668-76; and Skerra, 2008, *FEBSJ.,* 275:2677-83. In certain embodiments, phage or mammalian cell display technology may be used to produce and/or identify a VEGF/DLL4-binding polypeptide that is not an antibody. In certain embodiments, the polypeptide comprises a protein scaffold of a type selected from the group consisting of protein A, protein G, a lipocalin, a fibronectin domain, an ankyrin consensus repeat domain, and thioredoxin.

[00335] In certain embodiments, the VEGF/DLL4-binding agents or antibodies can be used in any one of a number of conjugated (i.e. an immunoconjugate or radioconjugate) or non-conjugated forms. In certain embodiments, the antibodies can be used in a non-conjugated form to harness the subject's natural defense mechanisms including complement-dependent cytotoxicity and antibody-dependent cellular toxicity to eliminate malignant or cancer cells.

[00336] In some embodiments, the VEGF/DLL4-binding agent (e.g., an antibody or polypeptide) is conjugated to a cytotoxic agent. In some embodiments, the cytotoxic agent is a chemotherapeutic agent including, but not limited to, methotrexate, adriamycin, doxorubicin, melphalan, mitomycin C, chlorambucil, daunorubicin or other intercalating agents. In some embodiments, the cytotoxic agent is an enzymatically active toxin of bacterial, fungal, plant, or animal origin, or fragments thereof, including, but not limited to, diphtheria A chain, non-binding active fragments of diphtheria toxin, exotoxin A chain, 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. In some embodiments, the cytotoxic agent is a radioisotope to produce a radioconjugate or a radioconjugated antibody. A variety of radionuclides are available for the production of radioconjugated antibodies including, but not limited to, ^{90}Y, ^{125}I, ^{131}I, ^{123}I, ^{111}In, ^{131}In, ^{105}Rh, ^{153}Sm, ^{67}Cu, ^{67}Ga, ^{166}Ho, ^{177}Lu, ^{186}Re, ^{188}Re and ^{212}Bi. Conjugates of an antibody and one or more small molecule toxins, such as calicheamicins, maytansine (e.g., mertansine), maytansinoid, trichothecene, and CC1065, and the derivatives of these toxins that have toxin activity, can also be used. Conjugates of an antibody and cytotoxic agent can be made using a variety of bifunctional protein-coupling agents including, but not limited to, N-succinimidyl-3-(2-pyridyidithiol) propionate (SPDP), iminothiolane (IT), bifunctional

derivatives of imidoesters (such as dimethyl adipimidate HCl), active esters (such as disuccinimidyl suberate), aldehydes (such as glutareldehyde), 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).

VI. Polynucleotides

[00337] In certain embodiments, the VEGF/DLL4 binding agents used in the present invention are encoded by one or more polynucleotides described herein. These polynucleotides can be polynucleotides that encode a polypeptide (or a fragment of a polypeptide) that specifically binds VEGF, DLL4, both VEGF and DLL4. The term "polynucleotides that encode a polypeptide" encompasses a polynucleotide which includes only coding sequences for the polypeptide, as well as a polynucleotide which includes additional coding and/or non-coding sequences. For example, in some embodiments, the polynucleotide comprises a polynucleotide sequence that encodes an antibody to human VEGF or encodes a fragment of such an antibody (e.g., a fragment comprising the antigen-binding site). In some embodiments, the polynucleotide comprises a polynucleotide sequence that encodes an antibody to human DLL4 or encodes a fragment of such an antibody (e.g., a fragment comprising the antigen-binding site). The polynucleotides can be in the form of RNA or in the form of DNA. DNA includes cDNA, genomic DNA, and synthetic DNA; and can be double-stranded or single-stranded, and if single-stranded can be the coding strand or non-coding (anti-sense) strand.

[00338] In certain embodiments, the polynucleotide comprises a polynucleotide encoding a polypeptide comprising a sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:56, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64. In certain embodiments, the polynucleotide comprises a polynucleotide encoding a polypeptide comprising a sequence selected from the group consisting of SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:56, SEQ ID NO:58, SEQ ID NO:62, and SEQ ID NO: 64. In some embodiments, the polynucleotide comprises a polynucleotide sequence selected from the group consisting of SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:40, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69, SEQ ID

NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, and SEQ ID NO:78.

[00339] In certain embodiments, the polynucleotide comprises a polynucleotide having a nucleotide sequence at least about 80% identical, at least about 85% identical, at least about 90% identical, at least about 95% identical, and in some embodiments, at least about 96%, 97%, 98% or 99% identical to a polynucleotide comprising a sequence selected from the group consisting of SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:55, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, and SEQ ID NO:78. In certain embodiments, the polynucleotide comprises a polynucleotide having a nucleotide sequence at least about 80% identical, at least about 85% identical, at least about 90% identical, at least about 95% identical, and in some embodiments, at least about 96%, 97%, 98% or 99% identical to a polynucleotide comprising a sequence selected from the group consisting of SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:40, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:54, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, and SEQ ID NO:78. Also provided is a polynucleotide that comprises a polynucleotide that hybridizes to SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:40, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, and SEQ ID NO:78. In certain embodiments, the hybridization is under conditions of high stringency.

[00340] In certain embodiments, the polynucleotides comprise the coding sequence for the mature polypeptide fused in the same reading frame to a polynucleotide which aids, for example, in expression and secretion of a polypeptide from a host cell (e.g., a leader sequence which functions as a secretory sequence for controlling transport of a polypeptide from the cell). The polypeptide having a leader sequence is a preprotein and can have the leader sequence cleaved by the host cell to form the mature form of the polypeptide. The polynucleotides can also encode for a proprotein which is the mature protein plus additional 5' amino acid residues. A mature protein having a prosequence is a proprotein and is an inactive form of the protein. Once the prosequence is cleaved an active mature protein remains.

[00341] In certain embodiments, the polynucleotides comprise the coding sequence for the mature polypeptide fused in the same reading frame to a marker sequence that allows, for example, for purification of the encoded polypeptide. For example, the marker sequence can be a hexa-histidine tag

supplied by a pQE-9 vector to provide for purification of the mature polypeptide fused to the marker in the case of a bacterial host, or the marker sequence can be a hemagglutinin (HA) tag derived from the influenza hemagglutinin protein when a mammalian host (e.g., COS-7 cells) is used. In some embodiments, the marker sequence is a FLAG-tag, a peptide of sequence DYKDDDDK (SEQ ID NO:45) which can be used in conjunction with other affinity tags.

[00342] The present invention further relates to variants of the hereinabove described polynucleotides encoding, for example, fragments, analogs, and/or derivatives.

[00343] In certain embodiments, the polynucleotides comprise polynucleotides having a nucleotide sequence at least about 80% identical, at least about 85% identical, at least about 90% identical, at least about 95% identical, and in some embodiments, at least about 96%, 97%, 98% or 99% identical to a polynucleotide encoding a polypeptide comprising a VEGF/DLL4-binding agent (e.g., an antibody), or fragment thereof, described herein.

[00344] As used herein, the phrase a polynucleotide having a nucleotide sequence at least, for example, 95% "identical" to a reference nucleotide sequence is intended to mean that the nucleotide sequence of the polynucleotide is identical to the reference sequence except that the polynucleotide sequence can include up to five point mutations per each 100 nucleotides of the reference nucleotide sequence. In other words, to obtain a polynucleotide having a nucleotide sequence at least 95% identical to a reference nucleotide sequence, up to 5% of the nucleotides in the reference sequence can be deleted or substituted with another nucleotide, or a number of nucleotides up to 5% of the total nucleotides in the reference sequence can be inserted into the reference sequence. These mutations of the reference sequence can occur at the 5' or 3' terminal positions of the reference nucleotide sequence or anywhere between those terminal positions, interspersed either individually among nucleotides in the reference sequence or in one or more contiguous groups within the reference sequence.

[00345] The polynucleotide variants can contain alterations in the coding regions, non-coding regions, or both. In some embodiments, a polynucleotide variant contains alterations which produce silent substitutions, additions, or deletions, but does not alter the properties or activities of the encoded polypeptide. In some embodiments, a polynucleotide variant comprises silent substitutions that results in no change to the amino acid sequence of the polypeptide (due to the degeneracy of the genetic code). Polynucleotide variants can be produced for a variety of reasons, for example, to optimize codon expression for a particular host (i.e., change codons in the human mRNA to those preferred by a bacterial host such as E. coli). In some embodiments, a polynucleotide variant comprises at least one silent mutation in a non-coding or a coding region of the sequence.

[00346] In some embodiments, a polynucleotide variant is produced to modulate or alter expression (or expression levels) of the encoded polypeptide. In some embodiments, a polynucleotide variant is produced to increase expression of the encoded polypeptide. In some embodiments, a polynucleotide

variant is produced to decrease expression of the encoded polypeptide. In some embodiments, a polynucleotide variant has increased expression of the encoded polypeptide as compared to a parental polynucleotide sequence. In some embodiments, a polynucleotide variant has decreased expression of the encoded polypeptide as compared to a parental polynucleotide sequence.

[00347] In some embodiments, at least one polynucleotide variant is produced (without changing the amino acid sequence of the encoded polypeptide) to increase production of a heteromultimeric molecule. In some embodiments, at least one polynucleotide variant is produced (without changing the amino acid sequence of the encoded polypeptide) to increase production of a bispecific antibody.

[00348] In certain embodiments, the polynucleotides are isolated. In certain embodiments, the polynucleotides are substantially pure.

[00349] Vectors and cells comprising the polynucleotides described herein are also provided. In some embodiments, an expression vector comprises a polynucleotide molecule. In some embodiments, a host cell comprises an expression vector comprising the polynucleotide molecule. In some embodiments, a host cell comprises a polynucleotide molecule.

VII. Kits comprising VEGF/DLL4-binding agents

[00350] The present invention also provides kits that comprise the VEGF/DLL4-binding agents (e.g., antibodies) and at least one additional therapeutic agent. Also provided are kits comprising a VEGF/DLL4-binding agent (e.g., an anti-VEGF/anti-DLL4 bispecific antibody such as 305B83), as well as at least one additional therapeutic agent. In certain embodiments, the second (or more) therapeutic agent is a chemotherapeutic agent. In certain embodiments, the second (or more) therapeutic agent is an angiogenesis inhibitor. In certain embodiments, the additional agent(s) are selected from the group consisting of (a) leucovorin, 5-fluorouracil, and irinotecan; (b) paclitaxel; (c) gemcitabine and ABRAXANE®; and (d) paclitaxel and carboplatin. The kits may be configured for any of the dosage regimens described herein.

[00351] Embodiments of the present disclosure can be further defined by reference to the following non-limiting examples, which describe in detail preparation of certain antibodies of the present disclosure and methods for using antibodies of the present disclosure. It will be apparent to those skilled in the art that many modifications, both to materials and methods, may be practiced without departing from the scope of the present disclosure.

EXAMPLES

Example 1

Clinical trial test 305B83 with paclitaxel in ovarian, primary peritoneal, or fallopian tube cancer

[00352] A Phase 1b study of paclitaxel plus OMP-305B83 in subjects with platinum resistant (defined as having progressed ≤ 6 months from the completion of therapy without progressing during the treatment) Grade 2 or 3 ovarian, primary peritoneal, or fallopian tube cancer is performed. Up to 30 subjects are enrolled in the study. Subjects must have received prior bevacizumab and/or more than 2 prior therapies. In addition, subjects must not have received prior weekly paclitaxel for recurrent disease. Prior to enrollment, subjects undergo screening to determine study eligibility. Dexamethasone, an antihistamine, and an H-2 blocker are given as premedications prior to administering paclitaxel. Paclitaxel 80 mg/m^2 is administered intravenously on Days 0, 7, and 14 of each 28 day cycle and is continued until confirmed complete response, intolerance, or disease progression. OMP-305B83 is administered prior to paclitaxel by intravenous (IV) infusion. In the dose escalation portion of the study, subjects will be dosed at 3, 5, and 10 mg/kg administered IV once every 2 weeks. No dose escalation or reduction of OMP-305B83 will be allowed within a dose cohort. Three subjects will be treated at each dose level if no dose-limiting toxicities (DLTs) are observed. If 1 of 3 subjects experiences a DLT, that dose level will be expanded to 6 subjects. If 2 or more subjects experience a DLT, no further subjects will be dosed at that level and 3 additional subjects will be added to the preceding dose cohort unless 6 subjects have already been treated at that dose level. Subjects will be assessed for DLTs from Days 0–28. Once the maximum tolerated dose of OMP-305B83 (i.e., either 3, 5, or 10 mg/kg once every 2 weeks) in combination with paclitaxel has been established, additional subjects will be enrolled in an expansion cohort, so that a total of 30 subjects will be treated in the study. Treatment will be continued until confirmed complete response, intolerance or disease progression.

[00353] Subjects are assessed for response at study Day 56. If a subject has not had progressive disease per the Response Evaluation Criteria in Solid Tumors (RECIST 1.1) criteria at the Study Day 56 response assessments, treatment may be continued at the subject's initial dose level on an every-two-week basis until disease progression occurs.

Example 2

Clinical trial testing 305B83 with paclitaxel and carboplatin in endometrial cancer

[00354] A Phase 1b clinical trial testing the combination of the anti-VEGF/DLL4 bispecific antibody 305B83 in combination with paclitaxel and carboplatin is performed as a first-line therapy in endometrial cancer (Figure 1B). A dose escalation starting at 1 mg/kg is performed. In this trial, three patients are initially dosed at a first level (1 mg/kg) every two weeks or every three weeks with 305B83 and the paclitaxel/carboplatin combination is administered as per standard of care. If no dose-limiting toxicity

(DLT) is observed in the patients at the first level, three additional patients will then be dosed at the second level (2.5 mg/kg). If a single patient exhibits DLT at the first level, then three additional subjects will be treated at the first level. If no other of the six patients at the first level experiences a DLT, then three additional patients will be dosed at the second level, and the process will be repeated at the second level. If two patients exhibit DLT, then no further subjects will be treated at the first level. In general, the maximum tolerated dose (MTD) will be the level at which 0-1 subjects experienced a DLT. The dosing level escalation continues at 2.5 mg/kg and 5.0 mg/kg.

[00355] Following dose escalation, an expansion cohort at the MTD is treated, and safety of the treatment is monitored, including physical examination, vital signs, laboratory test results including brain natriuretic peptide (BNP) levels, every 21 days. Patients can be treated until their disease progresses or unless other safety criteria suggest that discontinuation of therapy is appropriate.

Example 3

Clinical trial of 305B83 with FOLFIRI for colorectal cancer

[00356] A Phase 1b clinical trial testing the combination of the anti-VEGF/DLL4 bispecific antibody and FOLFIRI as a second-line therapy in colorectal cancer is performed (Figure 2A). As described above in Example 1 or 2, a dose escalation is performed at 3 mg/kg, 5 mg/kg, 10 mg/kg, and optionally 5 mg/kg given every two weeks or every three weeks followed by an expansion cohort at the MTD. Once an MTD is established, patients can be treated until their disease progresses or unless other safety criteria suggest that discontinuation of therapy is appropriate.

[00357] In another example, a Phase 1b clinical trial testing the combination of the anti-VEGF/DLL4 bispecific antibody and FOLFIRI as a second-line therapy in colorectal cancer is performed (Figure 2B). As described above in Example 2, a dose escalation is performed at 1 mg/kg, 2 mg/kg, 4 mg/kg, and optionally 5 mg/kg given every two weeks or every three weeks followed by an expansion cohort at the MTD. Once an MTD is established, patients can be treated until their disease progresses or unless other safety criteria suggest that discontinuation of therapy is appropriate.

Example 4

Clinical trial of 305B83 with gemcitabine and ABRAXANE® for pancreatic cancer

[00358] A Phase 1b clinical trial testing the combination of the anti-VEGF/DLL4 bispecific antibody with gemcitabine and ABRAXANE® as a first-line therapy in pancreatic cancer is performed (Figure 3). This study starts with a dose expansion starting at either 0.5 mg/kg or 1 mg/kg (as shown).

[00359] This is a Phase 1b dose-escalation study of ABRAXANE®, gemcitabine, and 305B83 in subjects with first line metastatic pancreatic cancer. Up to a total of 24 subjects are treated. Subjects are assessed for safety, immunogenicity, efficacy, and exploratory biomarkers. Prior to enrollment, subjects undergo screening to determine study eligibility. Patients then receive gemcitabine administered by intravenous (IV) infusion at a dose of 1000 mg/m^2 on Days 0, 7, and 14 of each 28-day treatment cycle (or until

toxicity necessitates reducing or holding a dose) and ABRAXANE® which is administered by IV infusion at a dose of 125 mg/m^2 over 30 minutes on Days 0, 7, and 14 of each 28-day treatment cycle.

[00360] 305B83 is administered by IV infusion once every 14 days or every 21 days over 30 minutes. In the initial phase of the study, dose escalation is conducted to determine the maximum tolerated dose (MTD). The dose levels of 305B83 will be 0.5, 1.0, 2, optionally 4, and optionally 5 mg/kg administered IV once every two or every three weeks. No dose escalation or reduction is allowed within a dose cohort. Intermediate dosing cohorts can be added upon agreement with the investigators and study sponsor. In addition, alternate dosing schedule cohorts of 305B83 (e.g., every four-week dosing) can be studied upon agreement with the investigators and the study sponsor. Three subjects are treated in each dose levels if no dose-limiting toxicities (DLTs) are observed. If one of three subjects experiences a DLT, that dose level is expanded to six subjects. If two or more subjects experience a DLT, no further subjects are dosed at that level, and three additional subjects are added to the preceding dose cohort, unless six subjects have already been treated at that dose level. Subjects are assessed for DLTs from the time of the first dose through Day 28. Dose escalation, if appropriate, occurs after all subjects in a cohort have completed their Day 28 DLT assessment. The maximum tolerated dose (MTD) is the highest dose level at which zero or one of six subjects experienced a DLT (i.e., six subjects will ultimately be treated at the MTD dose level). If the MTD is not been reached after the highest tested scheduled dose, then that dose (e.g., 2 mg/kg, 4 mg/kg, or 5 mg/kg) is considered the MTD. Following completion of the dose escalation portion of the study, six patients are enrolled in an expansion cohort and treated at the MTD.

[00361] Safety is assessed by adverse event monitoring (including attribution of adverse events and serious adverse events), physical examination, vital signs, clinical laboratory testing including assessment of BNP every 21 days, Doppler echocardiogram, anti-305B83 testing, urinalysis, and subject interview on an ongoing basis from enrollment through 30 days following the discontinuation of treatment. Any subject who has two consecutive BNP values >100 pg/ml or one value >200 pg/ml is started on an ACE inhibitor or carvedilol. Subjects are assessed for disease status every 8 weeks and for safety at every visit and through 30 days following treatment termination. Biomarker assessment is performed at Study Days 0, 21, 49, 70 then every 12 weeks and at treatment termination. Serum samples for PK and immunogenicity are also obtained.

Example 5

Simultaneous blockage of DLL4 and VEGF produces superior anti-tumor effects

[00362] Simultaneous inhibition of DLL4 and VEGF by 305B83 plus anti-mDLL4 and anti-mVEGF antibodies produced anti-tumor effects superior to that of anti-hDLL4 + anti-mDLL4 or anti-hVEGF + anti-mVEGF. Furthermore, the combination of DLL4 and VEGF inhibition induced significant down-regulation of vasculature-related genes and decreased vascular density in tumors, suggesting that the anti-VEGF-mediated inhibition of angiogenesis was dominant over the anti-DLL4 effect on endothelial cell

hyperproliferation. At doses where both anti-DLL4 and anti-VEGF alone produced suboptimal anti-tumor effect, dual targeting resulted in an enhanced tumor growth inhibition. 305B83 plus anti-mDLL4 and anti-mVEGF in combination with chemotherapeutic agents resulted in tumor shrinkage, and this effect was sustained after discontinuation of chemotherapy. In addition to the experiments using the anti-murine surrogate antibodies, a human skin graft model was used to evaluate the effect of anti-DLL4/VEGF on tumor- and stroma-derived VEGF and DLL4 where tumor cells grow in a human microenvironment.

[00363] These experiments were carried out by including surrogate antibodies – anti-mouse DLL4 (21R30) and anti-mouse VEGF (B20). Using a patient-derived ovarian xenograft tumor OMP-OV40, simultaneous inhibition of DLL4 and VEGF was shown to produce an anti-tumor effect superior to that of either anti-DLL4 or anti-VEGF alone (Figure 4). Simultaneous inhibition of DLL4 and VEGF by 305B83 plus mDLL4 and mVEGF induced significant down-regulation of vasculature-related genes and decreased vasculature density in tumor stroma, suggesting a dominant anti-VEGF-mediated effect resulting in inhibiting angiogenesis over the anti-DLL4 effect of endothelial cell hyperproliferation (Figure 5). 305B83 modulated Notch target gene expression in tumors similarly to anti-DLL4. Notably, at doses where both anti-DLL4 and anti-VEGF alone produced suboptimal anti-tumor effect, dual targeting resulted in an enhanced anti-tumor growth inhibition (Figure 6A). Similar results were observed in a gastric tumor OMP-STM1 (Figure 6B).

Example 6

Simultaneous blockage of DLL4 and VEGF delays tumor recurrence

[00364] To determine the effect of simultaneous blockade of DLL4 and VEGF by the bi-specific antibody 305B83 plus 21R30 (anti-mDLL4) and B20 (anti-mVEGF) on tumor recurrence, an ovarian serous carcinoma tumor model OMP-OV19 and a pancreatic adenocarcinoma model OMP-PN42 were treated in combination with standard of care agents (paclitaxel in ovarian cancer and gemcitabine/nab-paclitaxel in pancreatic cancer) for four weeks followed by a chemotherapy maintenance phase for 3-4 weeks. Subsequently, tumor growth was monitored for up to two months following the discontinuation of treatment. Our results showed that chemotherapy-treated tumors grew continuously during the course of study. The combination of chemotherapeutic agent with 305B83 plus 21R30 and B20 resulted in tumor shrinkage, and this effect was sustained after discontinuation of chemotherapy (Figures 7A and 7B). At the conclusion of the study, tumors were either completely regressed or stabilized.

Example 7

305B83 inhibits growth of tumors implanted into human skin transplants

[00365] To evaluate the effect of the bispecific anti-DLL4/VEGF antibody on tumor growth in a microenvironment composed of human cells, the human-mouse chimera skin graft model, in which human tumor cells are implanted intradermally into the full thickness of human skin previously

transplanted onto mice was used. The human microenvironment in this model provides both tumor- and stroma-derived VEGF and DLL4 targets, which allows us to evaluate in vivo efficacy. As shown in Figures 8A and 8B, 305B83 caused significant inhibition of tumor growth (87% TGI), compared to control antibody ($p<0.00005$), and this effect was superior to either demcizumab (45% TGI) or bevacizumab (70% TGI).

[00366] The studies described in the examples above demonstrate that simultaneous inhibition of DLL4 and VEGF produced anti-tumor effects superior to treatment with either anti-DLL4 or anti-VEGF alone. Simultaneous inhibition of DLL4 and VEGF induced significant down-regulation of vasculature-related genes and decreased vasculature density, suggesting a dominant anti-VEGF-mediated anti-angiogenic effect over anti-DLL4 mediated endothelial cell hyperproliferation. The combination of chemotherapeutic agents with anti-DLL4/VEGF resulted in tumor regression and significantly delayed tumor recurrence after treatment termination. In a human skin graft model, the bi-specific antibody produces a significant inhibition of colon tumor growth compared with either demcizumab or bevacizumab.

Example 8

305B83 is active in combination with gemcitabine in pancreatic cancer

[00367] In our initial experiment, the anti-tumor activity of anti-DLL4/VEGF to anti-DLL4 (demcizumab) and to anti-VEGF (bevacizumab) in combination with gemcitabine patient-derived pancreatic cancer xenografts was compared. Following four weeks of treatment, the gemcitabine treatment was discontinued and the antibody treatments were maintained. In the control group, tumors re-grew rapidly after gemcitabine was withdrawn, and the combination of gemcitabine with bevacizumab had no effect. In contrast, treatment with either demcizumab or the anti-DLL4/VEGF bispecific significantly delayed tumor growth (Figure 9).

[00368] To determine the effect of DLL4 and VEGF inhibition on tumor initiating cell frequency in pancreatic cancer, serial transplantation studies were performed. OMP-PN8 tumor-bearing mice were treated with a control Ab, demcizumab, bevacizumab, or the DLL4/VEGF bispecific. Following four weeks of treatment, the tumors were harvested, and tumor cells from each treatment were implanted into a new set of mice. Tumors were then allowed to grow for 83 days without treatment. In the control group, nine out of ten mice grew large tumors (Figure 10). The tumor growth frequency was reduced by anti-DLL4 treatment and the DLL4/VEGF bispecific, but not by bevacizumab, showing that the anti-CSC activity of demcizumab is retained in the DLL4/VEGF bispecific.

[00369] In patient-derived xenograft experiments, the stroma and vasculature is comprised of murine cells, whereas the tumor cells are human. Because DLL4 and VEGF are expressed in both tumor cells and in the stroma/vasculature, the previous experiments that were carried out with DLL4 and VEGF antagonists that block signaling of the human, but not murine, proteins and may have underestimated the full anti-tumor effect of blocking these pathways. To address this issue, the effect of simultaneous blockade of DLL4 and

VEGF in both human tumor and murine stroma/vasculature cells in our xenograft studies were evaluated. This was done with experiments including surrogate antibodies – anti-mouse DLL4 (21R30) and anti-mouse VEGF (B20) in addition to 305B83 (which blocks human DLL4 and VEGF). Complete DLL4/VEGF inhibition in combination with gemcitabine plus nab-paclitaxel in OMP-PN42 tumors was tested. As shown in Figure 11, gemcitabine treatment alone had minimal effect on this tumor. Including nab-paclitaxel delayed tumor growth, but eventually tumors grew in the group treated with this chemotherapy doublet. In contrast, the combination of DLL4/VEGF inhibition with gemcitabine and nab-paciltaxel treatment resulted in complete tumor regression.

Example 9

Management of hypertension in patients receiving 305B83

[00370] One of the side effects observed during the Phase 1a trial was development or exacerbation of hypertension in some patients receiving 305B83. To manage this side effect, a treatment algorithm was developed. First, if the patient's blood pressure exceeds 140/90, the patient is instructed to phone the principal investigator. The initial goal is to normalize blood pressure in 48-72 hours. Treatment with antihypertensive is to be adjusted daily if the blood pressure is not less than 140/90.

[00371] If the systolic pressure exceeds 180 mm Hg, then either hydralazine or clonidine is used to bring lower blood pressure rapidly. These agents are not to be used for either patient with systolic blood pressure lower than 180 mm Hg or for chronic hypertension management.

[00372] Prior to administering 305B83 and to maintain chronic blood pressure control, one of amlodipine and Procardia XL® is used as a first chronic anti-hypertension medication. The recommended starting dose for amlodipine is 5 mg orally daily. The dose should be adjusted daily if blood pressure is not controlled, until a maximum dose of 10 mg orally daily is reached. The recommended starting dose for Procardia XL® is 30-60 mg orally daily. The dose is adjusted daily if blood pressure is not controlled, until a maximum dose of 120 mg orally daily is reached.

[00373] If blood pressure is not controlled after the maximum efficacious dose of amlodipine or Procardia XL® has been given, then an angiotensin-converting-enzyme (ACE) inhibitor (if heart rate is low) or a beta blocker such as carvedilol (if heart rate is high) is added.

[00374] If the BP is not controlled on amlodipine or Procardia XL® plus the maximum dose of the second anti-hypertensive, a third anti-hypertensive is added to the mix. The third anti-hypertensive agent should be either an ACE inhibitor or beta blocker, whichever of these agents was not added as the second anti-hypertensive.

[00375] Patients already taking anti-hypertensive therapy at study entry should still follow this general algorithm, unless contraindicated. That is, they should be given a prescription for hydralazine or clonidine prior to dosing, if appropriate. In addition, if the patient is not already receiving amlodipine, Procardia

XL®, or a similar calcium channel blocker, amlodipine or Procardia XL® should be the first agent added to their existing anti-hypertensive regimen.

[00376] It is understood that the examples and embodiments described herein are for illustrative purposes only and that various modifications or changes in light thereof will be suggested to persons skilled in the art and are to be included within the spirit and purview of this application.

[00377] All publications, patents, patent applications, internet sites, and accession numbers/database sequences including both polynucleotide and polypeptide sequences cited herein are hereby incorporated by reference herein in their entirety for all purposes to the same extent as if each individual publication, patent, patent application, internet site, or accession number/database sequence was specifically and individually indicated to be so incorporated by reference.

[00378] Following are the sequences disclosed in the application:

21M18 Heavy chain with signal sequence (underlined) (SEQ ID NO:1)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAP
GQGLEWIGYISSYNGATNYNQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYD
YDVGMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKC
CVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
REPQVYTLPPSREEMTKNQVSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
FFLYSELTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

21R79 Heavy chain with signal sequence (underlined) (SEQ ID NO:2)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAP
GQGLEWIGYIANYNRATNYNQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYD
YDVGMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKC
CVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
REPQVYTLPPSREEMTKNQVSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
FFLYSELTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

219R45 Heavy chain with signal sequence (underlined) (SEQ ID NO:3)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKVSCKASGYTFTNYWMHWVRQAP
GQGLEWMGDINPSNGRTSYKEKFKRRVTLSVDKSSSTAYMELSSLRSEDTAVYFCTIHYD
DKYYPLMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVS
WNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVER
KCCVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDG
VEVHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKG
QPREPQVYTLPPSREKMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLKSD
GSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

Light chain with signal sequence (underlined) (SEQ ID NO:4)
MVLQTQVFISLLLWISGAYGDIVMTQSPDSLAVSLGERATISCRASESVDNYGISFMKWF

QQKPGQPPKLLIYAASNQGSGVPDRFSGSGSGTDFTLTISSLQAEDVAVYYCQQSKEVPW
TFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQS
GNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

21M18 Heavy chain without predicted signal sequence (SEQ ID NO:5)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYISSYNGATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSSA
STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
LYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFL
FPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRV
VSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQ
VSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSELTVDKSRWQQGNV
FSCSVMHEALHNHYTQKSLSLSPGK

21R79 Heavy chain without predicted signal sequence (SEQ ID NO:6)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYIANYNRATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSSA
STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
LYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFL
FPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRV
VSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQ
VSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSELTVDKSRWQQGNV
FSCSVMHEALHNHYTQKSLSLSPGK

219R45 Heavy chain without predicted signal sequence (SEQ ID NO:7)
QVQLVQSGAEVKKPGASVKVSCKASGYTFTNYWMHWVRQAPGQGLEWMGDINPSNGRTSY
KEKFKRRVTLSVDKSSSTAYMELSSLRSEDTAVYFCTIHYDDKYYPLMDYWGQGTLVTVS
SASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQS
SGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSV
FLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTF
RVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREKMTK
NQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLKSDGSFFLYSKLTVDKSRWQQG
NVFSCSVMHEALHNHYTQKSLSLSPGK

Light chain without predicted signal sequence (SEQ ID NO:8)
DIVMTQSPDSLAVSLGERATISCRASESVDNYGISFMKWFQQKPGQPPKLLIYAASNQGS
GVPDRFSGSGSGTDFTLTISSLQAEDVAVYYCQQSKEVPWTFGGGTKVEIKRTVAAPSVI
FPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSS
TLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

21M18 Heavy chain variable region (SEQ ID NO:9)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYISSYNGATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSS

21R79 Heavy chain variable region (SEQ ID NO:10)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYIANYNRATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSS

219R45 Heavy chain variable region (SEQ ID NO:11)
QVQLVQSGAEVKKPGASVKVSCKASGYTFTNYWMHWVRQAPGQGLEWMGDINPSNGRTSY
KEKFKRRVTLSVDKSSSTAYMELSSLRSEDTAVYFCTIHYDDKYYPLMDYWGQGTLVTVSS

Light chain variable region (SEQ ID NO:12)
DIVMTQSPDSLAVSLGERATISCRASESVDNYGISFMKWFQQKPGQPPKLLIYAASNQGS

GVPDRFSGSGSGTDFTLTISSLQAEDVAVYYCQQSKEVPWTFGGGTKVEIK

21R75, 21R79, 21R83, and 21M18 Heavy chain CDR1 (SEQ ID NO:13)
TAYYIH

21R79 Heavy chain CDR2 (SEQ ID NO:14)
YIANYNRATNYNQKFKG

21M18 Heavy chain CDR2 (SEQ ID NO:15)
YISSYNGATNYNQKFKG

21R75, 21R79, 21R83, and 21M18 Heavy chain CDR3 (SEQ ID NO:16)
RDYDYDVGMDY

219R45 Heavy chain CDR1 (SEQ ID NO:17)
NYWMH

219R45 Heavy chain CDR2 (SEQ ID NO:18)
DINPSNGRTSYKEKFKR

219R45 Heavy chain CDR3 (SEQ ID NO:19)
HYDDKYYPLMDY

Light chain CDR1 (SEQ ID NO:20)
RASESVDNYGISFMK

Light chain CDR2 (SEQ ID NO:21)
AASNQGS

Light chain CDR3 (SEQ ID NO:22)
QQSKEVPWTFGG

Human DLL4 with signal sequence (underlined) (SEQ ID NO:23)
MAAASRSASGWALLLLVALWQQRAAGSGVFQLQLQEFINERGVLASGRPCEPGCRTFFRV
CLKHFQAVVSPGPCTFGTVSTPVLGTNSFAVRDDSSGGGRNPLQLPFNFTWPGTFSLIIE
AWHAPGDDLRPEALPPDALISKIAIQGSLAVGQNWLLDEQTSTLTRLRYSYRVICSDNYY
GDNCSRLCKKRNDHFGHYVCQPDGNLSCLPGWTGEYCQQPICLSGCHEQNGYCSKPAECL
CRPGWQGRLCNECIPHNGCRHGTCSTPWQCTCDEGWGGLFCDQDLNYCTHHSPCKNGATC
SNSGQRSYTCTCRPGYTGVDCELELSECDSNPCRNGGSCKDQEDGYHCLCPPGYYGLHCE
HSTLSCADSPCFNGGSCRERNQGANYACECPPNFTGSNCEKKVDRCTSNPCANGGQCLNR
GPSRMCRCRPGFTGTYCELHVSDCARNPCAHGGTCHDLENGLMCTCPAGFSGRRCEVRTS
IDACASSPCFNRATCYTDLSTDTFVCNCPYGFVGSRCEFPVG

Human DLL4 without predicted signal sequence (SEQ ID NO:24)
SGVFQLQLQEFINERGVLASGRPCEPGCRTFFRVCLKHFQAVVSPGPCTFGTVSTPVLGT
NSFAVRDDSSGGGRNPLQLPFNFTWPGTFSLIIEAWHAPGDDLRPEALPPDALISKIAIQ
GSLAVGQNWLLDEQTSTLTRLRYSYRVICSDNYYGDNCSRLCKKRNDHFGHYVCQPDGNL
SCLPGWTGEYCQQPICLSGCHEQNGYCSKPAECLCRPGWQGRLCNECIPHNGCRHGTCST
PWQCTCDEGWGGLFCDQDLNYCTHHSPCKNGATCSNSGQRSYTCTCRPGYTGVDCELELS
ECDSNPCRNGGSCKDQEDGYHCLCPPGYYGLHCEHSTLSCADSPCFNGGSCRERNQGANY
ACECPPNFTGSNCEKKVDRCTSNPCANGGQCLNRGPSRMCRCRPGFTGTYCELHVSDCAR
NPCAHGGTCHDLENGLMCTCPAGFSGRRCEVRTSIDACASSPCFNRATCYTDLSTDTFVC
NCPYGFVGSRCEFPVG

Human DLL4 N-Terminal Region (SEQ ID NO:25)
SGVFQLQLQEFINERGVLASGRPCEPGCRTFFRVCLKHFQAVVSPGPCTFGTVSTPVLGT
NSFAVRDDSSGGGRNPLQLPFNFTWPGTFSLIIEAWHAPGDDLRPEALPPDALISKIAIQ
GSLAVGQN

Human DLL4 DSL Domain (SEQ ID NO:26)
WLLDEQTSTLTRLRYSYRVICSDNYYGDNCSRLCKKRNDHFGHYVCQPDGNLSCLPGWTG
EYC

Human VEGF-A with signal sequence (underlined) (SEQ ID NO:27)
MNFLLSWVHWSLALLLYLHHAKWSQAAPMAEGGGQNHHEVVKFMDVYQRSYCHPIETLVD
IFQEYPDEIEYIFKPSCVPLMRCGGCCNDEGLECVPTEESNITMQIMRIKPHQGQHIGEM
SFLQHNKCECRPKKDRARQEKKSVRGKGKGQKRKRKKSRYKSWSVYVGARCCLMPWSLPG
PHPCGPCSERRKHLFVQDPQTCKCSCKNTDSRCKARQLELNERTCRCDKPRR

Human VEGF-A without predicted signal sequence (SEQ ID NO:28)
APMAEGGGQNHHEVVKFMDVYQRSYCHPIETLVDIFQEYPDEIEYIFKPSCVPLMRCGGC
CNDEGLECVPTEESNITMQIMRIKPHQGQHIGEMSFLQHNKCECRPKKDRARQEKKSVRG
KGKGQKRKRKKSRYKSWSVYVGARCCLMPWSLPGPHPCGPCSERRKHLFVQDPQTCKCSC
KNTDSRCKARQLELNERTCRCDKPRR

21M18 Heavy chain nucleotide sequence (13B Version 1) (SEQ ID NO:29)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCTTACTACATCCACTGGGTCAAGCAGGCCCCT
GGGCAGGGCCTGGAATGGATCGGCTACATCTCCTCCTACAACGGCGCCACCAACTACAAC
CAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCCACCTCCACCGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC
TACGACGTGGGCATGGACTACTGGGGCCAGGGCACCCTGGTCACCGTGTCCTCTGCCTCC
ACCAAGGGCCCATCCGTGTTCCCTCTGGCCCCTTGCTCCCGGTCCACCTCTGAGTCTACC
GCCGCTCTGGGCTGCCTGGTGAAGGACTACTTCCCTGAGCCTGTGACCGTGTCCTGGAAC
TCTGGCGCCCTGACCTCTGGCGTGCACACCTTCCCTGCCGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCTAGCGTGGTGACCGTGCCTTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCTTCCAACACCAAGGTGGACAAGACCGTGGAGCGGAAGTGC
TGCGTGGAGTGCCCTCCTTGTCCTGCTCCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAGCCTAAGGACACCCTGATGATCTCCCGGACCCCTGAAGTGACCTGCGTGGTG
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAG
GTGCACAACGCCAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCTGTGCTGACCGTGGTGCACCAGGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGATATCGCCGTGGAGTGGGAGTCT
AACGGCCAGCCTGAGAACAACTACAAGACCACCCCTCCTATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
TCTCCTGGCAAGTAG

21R79 Heavy chain nucleotide sequence (13B Version 1) (SEQ ID NO:30)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTGAAACAGGCACCA
GGCCAGGGACTGGAATGGATCGGCTATATCGCCAACTACAACCGGGCCACCAACTACAAC
CAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCCACCTCCACAGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC

TACGACGTGGGCATGGACTACTGGGGCCAGGGCACCCTGGTGACAGTGTCCTCCGCCTCC
ACCAAGGGCCCCTCCGTGTTCCCTCTGGCCCCTTGCTCCCGGTCCACCTCTGAGTCTACC
GCCGCTCTGGGCTGCCTGGTGAAGGACTACTTCCCTGAGCCTGTGACCGTGTCCTGGAAC
TCTGGCGCCCTGACCTCTGGCGTGCACACCTTCCCTGCCGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCTAGCGTGGTGACCGTGCCTTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCTTCCAACACCAAGGTGGACAAGACCGTGGAGCGGAAGTGC
TGCGTGGAGTGCCCTCCTTGTCCTGCTCCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAGCCTAAGGACACCCTGATGATCTCCCGGACCCCTGAAGTGACCTGCGTGGTG
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAG
GTGCACAACGCCAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCTGTGCTGACCGTGGTGCACCAGGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGATATCGCCGTGGAGTGGGAGTCT
AACGGCCAGCCTGAGAACAACTACAAGACCACCCCTCCTATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
TCTCCTGGCAAGTAG

21R79 Heavy chain nucleotide sequence (13B Version 2) (SEQ ID NO:31)

ATGAAGCACCTATGGTTCTTTCTATTATTAGTGGCCGCTCCCCGTTGGGTGTTATCGCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCGCTAATTATAATAGAGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC
TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA
TCTCCTGGCAAGTAG

219R45 Heavy chain nucleotide sequence (13A Version 1 (SEQ ID NO:32)

ATGAAGCATCTGTGGTTTTTTCCTGTTGCTCGTGGCGGCACCCAGATGGGTGTTGTCCCAA
GTGCAGCTGGTCCAGAGCGGGGCTGAGGTGAAGAAACCCGGAGCAAGCGTAAAAGTATCG
TGTAAGGCCTCGGGGTACACGTTTACAAACTACTGGATGCATTGGGTGCGGCAGGCTCCG
GGACAGGGGTTGGAATGGATGGGTGACATTAACCCCTCAAATGGCAGAACATCATATAAG
GAAAAGTTCAAACGCCGCGTCACACTCTCCGTGGACAAGTCAAGCTCGACTGCGTACATG
GAACTTTCGTCGCTGAGGTCGGAGGACACGGCAGTGTACTTTTGCACCATCCATTATGAT
GACAAGTATTACCCTCTGATGGATTATTGGGGTCAGGGTACGTTGGTCACCGTCTCCAGC
GCGTCGACGAAAGGTCCCTCGGTATTTCCCCTCGCCCCCTGCTCGAGGTCGACATCCGAA

TCAACAGCTGCCCTCGGCTGCCTGGTCAAAGACTACTTCCCAGAGCCGGTAACGGTGTCG
TGGAACTCGGGAGCGCTTACGTCCGGAGTCCACACATTTCCGGCGGTACTGCAATCCTCG
GGACTGTATTCGTTGTCGTCAGTGGTGACTGTCCCGTCCTCCAATTTCGGGACTCAGACC
TATACGTGCAACGTCGACCACAAACCCTCAAACACCAAGGTGGATAAGACAGTGGAGCGC
AAGTGCTGCGTGGAGTGTCCCCCGTGTCCGGCACCCCCTGTCGCCGGACCCTCAGTCTTT
TTGTTTCCGCCGAAGCCCAAAGATACACTCATGATCTCAAGAACGCCCGAGGTAACATGC
GTGGTGGTCGATGTAAGCCACGAGGATCCAGAAGTACAATTCAATTGGTATGTAGACGGG
GTCGAGGTCCATAACGCAAAGACGAAACCGAGGGAAGAGCAGTTCAATTCGACTTTCCGG
GTGGTGTCGGTGCTTACAGTCGTACATCAGGACTGGTTGAACGGGAAGGAGTACAAGTGT
AAAGTATCGAATAAGGGCCTTCCAGCGCCGATTGAAAAGACCATCTCCAAGACCAAAGGA
CAGCCACGAGAGCCGCAAGTCTATACGCTTCCTCCCAGCCGAGAAAAGATGACTAAAAAC
CAGGTATCGCTTACGTGTCTCGTCAAGGGTTTCTACCCTTCGGACATCGCGGTGGAATGG
GAGAGCAATGGACAACCGGAAAACAACTACAAGACGACACCGCCTATGTTGAAAAGCGAT
GGATCGTTTTTCCTCTATTCGAAACTCACGGTCGATAAGTCACGGTGGCAGCAGGGGAAT
GTGTTCTCCTGTTCAGTGATGCACGAGGCGCTCCACAATCACTATACCCAGAAAAGCCTG
TCACTTTCCCCGGGAAAATGA

219R45 Heavy chain nucleotide sequence (13A Version 2) (SEQ ID NO:33)

ATGAAGCACCTCTGGTTCTTCCTGCTCCTCGTGGCTGCTCCTCGGTGGGTCCTCTCCCAA
GTGCAGCTGGTCCAGAGCGGGGCTGAGGTGAAGAAACCCGGAGCTTCCGTCAAAGTCTCC
TGTAAGGCTTCCGGATACACCTTTACCAACTATTGGATGCACTGGGTGCGGCAGGCTCCT
GGACAAGGGCTGGAATGGATGGGAGACATCAATCCTTCCAATGGCAGAACCTCCTACAAG
GAAAAATTCAAACGGCGGGTCACACTCTCCGTGGACAAGTCTAGCTCCACAGCTTACATG
GAACTCTCCTCCCTGCGGTCCGAAGACACAGCTGTCTACTTCTGCACCATCCACTACGAC
GACAAGTACTACCCTCTGATGGACTACTGGGGCCAGGGAACCCTGGTCACCGTGTCCAGC
GCTTCCACAAAAGGACCCTCCGTCTTTCCCCTCGCCCCCTGCTCCCGGTCCACATCCGAA
TCAACAGCTGCCCTCGGCTGCCTGGTCAAAGACTACTTCCCAGAGCCTGTCACAGTGTCC
TGGAACTCCGGAGCTCTCACATCCGGAGTCCACACATTTCCTGCTGTGCTCCAATCCTCC
GGACTGTATTCCCTCTCCTCCGTGGTGACAGTGCCTTCCTCCAATTTCGGGACACAGACC
TATACATGCAACGTGGACCACAAACCCTCCAACACCAAAGTCGATAAGACAGTGGAGCGC
AAGTGCTGCGTGGAGTGTCCCCCTTGTCCTGCTCCCCCTGTGGCTGGACCTTCCGTCTTT
CTGTTTCCTCCTAAACCTAAAGACACCCTCATGATCTCCCGGACCCCCGAGGTCACATGC
GTGGTCGTCGATGTGAGCCACGAGGACCCCGAAGTCCAATTTAATTGGTATGTGGACGGG
GTGGAGGTCCATAACGCTAAGACCAAACCTAGGGAAGAGCAGTTCAATTCCACTTTCCGG
GTGGTGTCCGTGCTGACCGTCGTTCATCAGGACTGGCTCAACGGGAAAGAATACAAATGC
AAAGTCTCTAATAAGGGCCTCCCTGCTCCTATTGAAAAAACAATTTCCAAAACAAAAGGA
CAACCTCGGGAGCCTCAAGTCTACACACTGCCACCTTCCCGGGAAAAAATGACAAAAAAT
CAAGTCTCCCTCACATGTCTCGTCAAGGGATTCTACCCTTCCGACATTGCTGTGGAATGG
GAATCCAATGGACAACCTGAAAACAACTACAAGACAACACCTCCTATGCTCAAAAGCGAT
GGGTCCTTTTTCCTCTATTCCAAACTCACAGTCGATAAGTCTCGGTGGCAGCAGGGGAAT
GTGTTCTCCTGTTCCGTGATGCACGAGGCTCTCCACAATCACTATACCCAGAAAAGCCTG
TCCCTCTCCCCTGGAAAATGA

Light chain nucleotide sequence (SEQ ID NO:34)

ATGGTGCTGCAGACCCAGGTGTTCATCTCCCTGCTGCTGTGGATCTCCGGCGCCTACGGC
GACATCGTGATGACCCAGTCCCCAGACTCCCTGGCTGTGTCTCTGGGAGAGCGGGCCACC
ATCTCTTGCAGAGCCTCCGAGTCCGTGGACAACTACGGCATCTCCTTCATGAAGTGGTTC
CAGCAGAAGCCCGGCCAGCCCCCAAAGCTGCTGATCTACGCCGCCTCCAACCAGGGATCT
GGCGTGCCCGACCGGTTCTCTGGATCCGGCTCTGGCACCGACTTTACCCTGACCATCAGC
TCCCTGCAGGCCGAGGACGTGGCCGTGTACTACTGCCAGCAGTCCAAAGAGGTGCCCTGG
ACCTTCGGCGGAGGCACCAAGGTGGAAATCAAGCGGACCGTGGCCGCTCCCTCCGTGTTC
ATCTTCCCACCCTCCGACGAGCAGCTGAAGTCCGGAACCGCCTCCGTCGTGTGCCTGCTG
AACAACTTCTACCCCCGCGAGGCCAAGGTGCAGTGGAAGGTGGACAACGCCCTGCAGTCC
GGCAACTCCCAGGAATCCGTCACCGAGCAGGACTCCAAGGACAGCACCTACTCCCTGTCC

TCCACCCTGACCCTGTCCAAGGCCGACTACGAGAAGCACAAGGTGTACGCCTGCGAAGTG
ACCCACCAGGGCCTGTCCAGCCCCGTGACCAAGTCCTTCAACCGGGGCGAGTGTTAG

21M18 Heavy chain variable region nucleotide sequence (SEQ ID NO:35)
CAGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATC
TCCTGCAAGGCCTCCGGCTACTCCTTCACCGCTTACTACATCCACTGGGTCAAGCAGGCC
CCTGGGCAGGGCCTGGAATGGATCGGCTACATCTCCTCCTACAACGGCGCCACCAACTAC
AACCAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCCACCTCCACCGCCTAC
ATGGAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTAC
GACTACGACGTGGGCATGGACTACTGGGGCCAGGGCACCCTGGTCACCGTGTCCTCT

21R79 Heavy chain variable region nucleotide sequence (13B) (SEQ ID NO:36)
CAGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATC
TCCTGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTGAAACAGGCA
CCAGGCCAGGGACTGGAATGGATCGGCTATATCGCCAACTACAACCGGGCCACCAACTAC
AACCAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCCACCTCCACAGCCTAC
ATGGAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTAC
GACTACGACGTGGGCATGGACTACTGGGGCCAGGGCACCCTGGTGACAGTGTCCTCC

21R79 Heavy chain variable region nucleotide sequence (13B Version 2) (SEQ ID NO:37)
CAGGTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATA
AGTTGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCA
CCAGGACAGGGACTTGAATGGATCGGATATATCGCTAATTATAATAGAGCTACAAACTAT
AACCAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATAC
ATGGAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTAT
GATTATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCT

219R45 Heavy chain variable region nucleotide sequence (13A version 1) (SEQ ID NO:38)
CAAGTGCAGCTGGTCCAGAGCGGGGCTGAGGTGAAGAAACCCGGAGCAAGCGTAAAAGTA
TCGTGTAAGGCCTCGGGGTACACGTTTACAAACTACTGGATGCATTGGGTGCGGCAGGCT
CCGGGACAGGGGTTGGAATGGATGGGTGACATTAACCCCTCAAATGGCAGAACATCATAT
AAGGAAAAGTTCAAACGCCGCGTCACACTCTCCGTGGACAAGTCAAGCTCGACTGCGTAC
ATGGAACTTTCGTCGCTGAGGTCGGAGGACACGGCAGTGTACTTTTGCACCATCCATTAT
GATGACAAGTATTACCCTCTGATGGATTATTGGGGTCAGGGTACGTTGGTCACCGTCTCC
AGC

219R45 Heavy chain variable region nucleotide sequence (13A Version 2) (SEQ ID NO:39)
CAAGTGCAGCTGGTCCAGAGCGGGGCTGAGGTGAAGAAACCCGGAGCTTCCGTCAAAGTC
TCCTGTAAGGCTTCCGGATACACCTTTACCAACTATTGGATGCACTGGGTGCGGCAGGCT
CCTGGACAAGGGCTGGAATGGATGGGAGACATCAATCCTTCCAATGGCAGAACCTCCTAC
AAGGAAAAATTCAAACGGCGGGTCACACTCTCCGTGGACAAGTCTAGCTCCACAGCTTAC
ATGGAACTCTCCTCCCTGCGGTCCGAAGACACAGCTGTCTACTTCTGCACCATCCACTAC
GACGACAAGTACTACCCTCTGATGGACTACTGGGGCCAGGGAACCCTGGTCACCGTGTCC
AGC

Light chain variable region nucleotide sequence (SEQ ID NO:40)
GACATCGTGATGACCCAGTCCCCAGACTCCCTGGCTGTGTCTCTGGGAGAGCGGGCCACC
ATCTCTTGCAGAGCCTCCGAGTCCGTGGACAACTACGGCATCTCCTTCATGAAGTGGTTC
CAGCAGAAGCCCGGCCAGCCCCCAAAGCTGCTGATCTACGCCGCCTCCAACCAGGGATCT
GGCGTGCCCGACCGGTTCTCTGGATCCGGCTCTGGCACCGACTTTACCCTGACCATCAGC
TCCCTGCAGGCCGAGGACGTGGCCGTGTACTACTGCCAGCAGTCCAAAGAGGTGCCCTGG
ACCTTCGGCGGAGGCACCAAGGTGGAAATCAAG

Human IgG1 Heavy chain constant region (SEQ ID NO:41)

ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSS
GLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCDKTHTCPPCPAPELLGG
PSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYN
STYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDE
LTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTVDKSRW
QQGNVFSCSVMHEALHNHYTQKSLSLSPGK

Human IgG2 Heavy chain constant region (SEQ ID NO:42)
ASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSS
GLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVF
LFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFR
VVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKN
QVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTVDKSRWQQGN
VFSCSVMHEALHNHYTQKSLSLSPGK

Human IgG3 Heavy chain constant region (SEQ ID NO:43)
ASTKGPSVFPLAPCSRSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSS
GLYSLSSVVTVPSSSLGTQTYTCNVNHKPSNTKVDKRVELKTPLGDTTHTCPRCPEPKSC
DTPPPCPRCPEPKSCDTPPPCPRCPEPKSCDTPPPCPRCPAPELLGGPSVFLFPPKPKDT
LMISRTPEVTCVVVDVSHEDPEVQFKWYVDGVEVHNAKTKPREEQYNSTFRVVSVLTVLH
QDWLNGKEYKCKVSNKALPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQVSLTCLVK
GFYPSDIAVEWESSGQPENNYNTTPPMLDSDGSFFLYSKLTVDKSRWQQGNIFSCSVMHE
ALHNRFTQKSLSLSPGK

Human IgG4 Heavy chain constant region (SEQ ID NO:44)
ASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSS
GLYSLSSVVTVPSSSLGTKTYTCNVDHKPSNTKVDKRVESKYGPPCPSCPAPEFLGGPSV
FLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTY
RVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTLPPSQEEMTK
NQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSRLTVDKSRWQEG
NVFSCSVMHEALHNHYTQKSLSLSLGK

FLAG peptide (SEQ ID NO:45)
DYKDDDDK

Parental 21R79 Heavy chain with signal sequence underlined unmodified chain (SEQ ID NO:46)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAP
GQGLEWIGYIANYNRATNYNQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYD
YDVGMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKC
CVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
REPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
FFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

Parental 219R45 Heavy chain with signal sequence underlined (SEQ ID NO:47)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKVSCKASGYTFTNYWMHWVRQAP
GQGLEWMGDINPSNGRTSYKEKFKRRVTLSVDKSSSTAYMELSSLRSEDTAVYFCTIHYD
DKYYPLMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVS
WNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVER
KCCVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDG
VEVHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKG
QPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSD
GSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

Parental 21R79 Heavy chain without predicted signal sequence (SEQ ID NO:48)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYIANYNRATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSSA
STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
LYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFL
FPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRV
VSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQ
VSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTVDKSRWQQGNV
FSCSVMHEALHNHYTQKSLSLSPGK

Parental 219R45 Heavy chain without signal sequence (SEQ ID NO:49)
QVQLVQSGAEVKKPGASVKVSCKASGYTFTNYWMHWVRQAPGQGLEWMGDINPSNGRTSY
KEKFKRRVTLSVDKSSSTAYMELSSLRSEDTAVYFCTIHYDDKYYPLMDYWGQGTLVTVS
SASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQS
SGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSV
FLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTF
RVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTK
NQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSKLTVDKSRWQQG
NVFSCSVMHEALHNHYTQKSLSLSPGK

Parental 21R79 Heavy chain variable region nucleotide sequence (SEQ ID NO:50)
CAAGTGCAGCTCGTGCAGTCAGGGGCGGAGGTCAAGAAGCCGGGAGCATCGGTCAAAATC
TCGTGTAAGGCCTCGGGGTACTCCTTTACTGCGTATTACATCCATTGGGTAAAGCAGGCG
CCAGGGCAGGGATTGGAGTGGATTGGGTATATCGCCAATTACAATCGCGCGACGAACTAT
AACCAGAAATTCAAGGGAAGGGTGACCTTCACAACGGATACATCGACATCGACGGCCTAC
ATGGAACTTCGCAGCCTGCGATCAGATGACACGGCGGTATACTATTGCGCAAGAGATTAC
GACTATGATGTGGGAATGGACTATTGGGGTCAAGGTACTCTGGTCACAGTCTCCTCC

Parental 219R45 Heavy chain variable region nucleotide sequence (SEQ ID NO:51)
CAGGTACAGCTCGTGCAATCGGGGGCAGAGGTCAAAAAGCCCGGTGCGTCGGTAAAGGTC
AGCTGCAAAGCGTCAGGTTATACATTCACGAATTACTGGATGCATTGGGTCAGACAGGCC
CCTGGACAAGGGCTTGAATGGATGGGAGATATCAATCCGTCGAACGGACGGACTAGCTAT
AAGGAGAAGTTTAAGAGGCGCGTAACACTGTCGGTGGACAAATCGTCCTCAACGGCCTAC
ATGGAGTTGTCATCCCTGCGGTCGGAAGATACGGCGGTCTACTTCTGTACTATCCACTAT
GACGATAAGTACTACCCGCTTATGGACTACTGGGGTCAGGGAACATTGGTAACCGTGAGC
AGC

Parental 21R79 Heavy chain nucleotide sequence with signal sequence (SEQ ID NO:52)
ATGAAACACTTGTGGTTTTTCCTCTTGCTCGTGGCAGCTCCTCGGTGGGTACTTTCACAA
GTGCAGCTCGTGCAGTCAGGGGCGGAGGTCAAGAAGCCGGGAGCATCGGTCAAAATCTCG
TGTAAGGCCTCGGGGTACTCCTTTACTGCGTATTACATCCATTGGGTAAAGCAGGCGCCA
GGGCAGGGATTGGAGTGGATTGGGTATATCGCCAATTACAATCGCGCGACGAACTATAAC
CAGAAATTCAAGGGAAGGGTGACCTTCACAACGGATACATCGACATCGACGGCCTACATG
GAACTTCGCAGCCTGCGATCAGATGACACGGCGGTATACTATTGCGCAAGAGATTACGAC
TATGATGTGGGAATGGACTATTGGGGTCAAGGTACTCTGGTCACAGTCTCCTCCGCCAGC
ACCAAGGGCCCTAGCGTCTTCCCTCTGGCTCCCTGCAGCAGGAGCACCAGCGAGAGCACA
GCCGCCCTGGGCTGCCTGGTCAAGGACTACTTCCCCGAACCGGTGACGGTGTCGTGGAAC
TCAGGCGCTCTGACCAGCGGCGTGCACACCTTCCCAGCTGTCCTACAGTCCTCAGGACTC
TACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAACTTCGGCACCCAGACCTACACC
TGCAACGTAGATCACAAGCCCAGCAACACCAAGGTGGACAAGACAGTTGAGCGCAAATGT
TGTGTCGAGTGCCCACCGTGCCCAGCACCACCTGTGGCAGGACCGTCAGTCTTCCTCTTC
CCCCCAAAACCCAAGGACACCCTCATGATCTCCCGGACCCCTGAGGTCACGTGCGTGGTG
GTGGACGTGAGCCACGAAGACCCCGAGGTCCAGTTCAACTGGTACGTGGACGGCGTGGAG

GTGCATAATGCCAAGACAAAGCCACGGGAGGAGCAGTTCAACAGCACGTTCCGTGTGGTC
AGCGTCCTCACCGTTGTGCACCAGGACTGGCTGAACGGCAAGGAGTACAAGTGCAAGGTC
TCCAACAAAGGCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAACCAAAGGGCAGCCC
CGAGAACCACAGGTGTACACCCTGCCCCCATCCCGGGAGGAGATGACCAAGAACCAGGTC
AGCCTGACCTGCCTGGTCAAAGGCTTCTACCCCAGCGACATCGCCGTGGAGTGGGAGAGC
AATGGGCAGCCGGAGAACAACTACAAGACCACACCTCCCATGCTGGACTCCGACGGCTCC
TTCTTCCTCTACAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAACGTCTTC
TCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTG
TCTCCGGGTAAA

Parental 219R45 Heavy chain nucleotide sequence with signal sequence (SEQ ID NO:53)
ATGAAACACCTCTGGTTCTTTTTGCTCCTGGTGGCAGCTCCCCGATGGGTGCTTAGCCAG
GTACAGCTCGTGCAATCGGGGGCAGAGGTCAAAAAGCCCGGTGCGTCGGTAAAGGTCAGC
TGCAAAGCGTCAGGTTATACATTCACGAATTACTGGATGCATTGGGTCAGACAGGCCCCT
GGACAAGGGCTTGAATGGATGGGAGATATCAATCCGTCGAACGGACGGACTAGCTATAAG
GAGAAGTTTAAGAGGCGCGTAACACTGTCGGTGGACAAATCGTCCTCAACGGCCTACATG
GAGTTGTCATCCCTGCGGTCGGAAGATACGGCGGTCTACTTCTGTACTATCCACTATGAC
GATAAGTACTACCCGCTTATGGACTACTGGGGTCAGGGAACATTGGTAACCGTGAGCAGC
GCGTCCACAAAGGGCCCTAGCGTCTTCCCTCTGGCTCCCTGCAGCAGGAGCACCAGCGAG
AGCACAGCCGCCCTGGGCTGCCTGGTCAAGGACTACTTCCCCGAACCGGTGACGGTGTCG
TGGAACTCAGGCGCTCTGACCAGCGGCGTGCACACCTTCCCAGCTGTCCTACAGTCCTCA
GGACTCTACTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAACTTCGGCACCCAGACC
TACACCTGCAACGTAGATCACAAGCCCAGCAACACCAAGGTGGACAAGACAGTTGAGCGC
AAATGTTGTGTCGAGTGCCCACCGTGCCCAGCACCACCTGTGGCAGGACCGTCAGTCTTC
CTCTTCCCCCCAAAACCCAAGGACACCCTCATGATCTCCCGGACCCCTGAGGTCACGTGC
GTGGTGGTGGACGTGAGCCACGAAGACCCCGAGGTCCAGTTCAACTGGTACGTGGACGGC
GTGGAGGTGCATAATGCCAAGACAAAGCCACGGGAGGAGCAGTTCAACAGCACGTTCCGT
GTGGTCAGCGTCCTCACCGTTGTGCACCAGGACTGGCTGAACGGCAAGGAGTACAAGTGC
AAGGTCTCCAACAAAGGCCTCCCAGCCCCCATCGAGAAAACCATCTCCAAAACCAAAGGG
CAGCCCCGAGAACCACAGGTGTACACCCTGCCCCCATCCCGGGAGGAGATGACCAAGAAC
CAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTACCCCAGCGACATCGCCGTGGAGTGG
GAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACACCTCCCATGCTGGACTCCGAC
GGCTCCTTCTTCCTCTACAGCAAGCTCACCGTGGACAAGAGCAGGTGGCAGCAGGGGAAC
GTCTTCTCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTC
TCCCTGTCTCCGGGTAAA

Parental 21R79 and 219R45 light chain variable region nucleotide sequence (SEQ ID NO:54)
GACATCGTGATGACCCAGTCCCCTGACTCCCTGGCTGTGTCCCTGGGCGAGAGGGCCACC
ATCTCCTGCAGAGCCAGCGAATCCGTCGATAATTATGGCATTTCCTTTATGAAGTGGTTC
CAGCAGAAACCAGGACAGCCTCCTAAGCTGCTCATTTACGCTGCATCCAACCAAGGGTCC
GGGGTCCCTGACAGGTTCTCCGGCAGCGGGTCCGGAACAGATTTCACTCTCACCATCAGC
AGCCTGCAGGCTGAAGATGTGGCTGTCTATTACTGTCAGCAAAGCAAGGAGGTGCCTTGG
ACATTCGGAGGAGGGACCAAGGTGGAAATCAAA

Parental 21R79 and 219R45 light chain nucleotide sequence (SEQ ID NO:55)
ATGGTGCTCCAGACCCAGGTCTTCATTTCCCTGCTGCTCTGGATCAGCGGAGCCTACGGG
GACATCGTGATGACCCAGTCCCCTGACTCCCTGGCTGTGTCCCTGGGCGAGAGGGCCACC
ATCTCCTGCAGAGCCAGCGAATCCGTCGATAATTATGGCATTTCCTTTATGAAGTGGTTC
CAGCAGAAACCAGGACAGCCTCCTAAGCTGCTCATTTACGCTGCATCCAACCAAGGGTCC
GGGGTCCCTGACAGGTTCTCCGGCAGCGGGTCCGGAACAGATTTCACTCTCACCATCAGC
AGCCTGCAGGCTGAAGATGTGGCTGTCTATTACTGTCAGCAAAGCAAGGAGGTGCCTTGG
ACATTCGGAGGAGGGACCAAGGTGGAAATCAAACGTACGGTGGCTGCCCCCTCCGTCTTC
ATCTTCCCCCCCAGCGATGAGCAGCTGAAAAGCGGCACTGCCAGCGTGGTGTGCCTGCTG
AATAACTTCTATCCCCGGGAGGCCAAAGTGCAGTGGAAGGTGGATAACGCCCTCCAAAGC

GGCAACTCCCAGGAGAGCGTCACAGAGCAGGACAGCAAGGACAGCACCTACAGCCTCAGC
AGCACCCTGACCCTGAGCAAAGCCGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTC
ACCCATCAGGGCCTGAGCAGCCCCGTCACAAAGAGCTTCAACAGGGGCGAGTGTTGA

21R75 Heavy chain without predicted signal sequence (SEQ ID NO:56)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYIAGYKDATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSSA
STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
LYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFL
FPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRV
VSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQ
VSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSELTVDKSRWQQGNV
FSCSVMHEALHNHYTQKSLSLSPGK

21R75 Heavy chain with predicted signal sequence (underlined) (SEQ ID NO:57)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAP
GQGLEWIGYIAGYKDATNYNQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYD
YDVGMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKC
CVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
REPQVYTLPPSREEMTKNQVSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
FFLYSELTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

21R75 Heavy chain variable region (SEQ ID NO:58)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYIAGYKDATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSS

21R75 Heavy chain CDR2 (SEQ ID NO:59)
YIAGYKDATNYNQKFKG

21R75 Heavy chain nucleotide sequence with signal sequence (13B Version 1) (SEQ ID NO:60)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCCCCT
GGACAGGGCCTGGAATGGATCGGCTATATCGCCGGCTACAAGGACGCCACCAACTACAAC
CAGAAATTCAAGGGCAGAGTGACCTTCACCACCGACACCTCCACCTCTACCGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC
TACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCCTCTGCTTCC
ACCAAGGGCCCCTCCGTGTTTCCTCTGGCCCCTTGCTCCAGATCCACCTCCGAGTCTACC
GCCGCTCTGGGCTGCCTCGTGAAGGACTACTTCCCCGAGCCCGTGACAGTGTCTTGGAAC
TCTGGCGCCCTGACCTCCGGCGTGCACACCTTTCCAGCTGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCCTCCGTCGTGACTGTGCCCTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCCTCCAACACCAAGGTGGACAAGACCGTGGAACGGAAGTGC
TGCGTGGAATGCCCCCCTTGTCCTGCCCCTCCTGTGGCTGGCCCTAGCGTGTTCCTGTTC
CCCCCAAAGCCCAAGGACACCCTGATGATCTCCCGGACCCCCGAAGTGACCTGCGTGGTG
GTGGATGTGTCCCACGAGGACCCCGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAA
GTGCACAACGCCAAGACCAAGCCCAGAGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCCGTGCTGACCGTGGTGCATCAGGACTGGCTGAACGGCAAAGAGTACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCCATCGAAAAGACCATCTCTAAGACCAAGGGACAGCCC
CGCGAGCCCCAGGTGTACACACTGCCTCCATCCCGGGAAGAGATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAAGGCTTCTACCCCTCCGATATCGCCGTGGAATGGGAGTCC
AACGGCCAGCCCGAGAACAACTACAAGACCACCCCCCCCCATGCTGGACTCCGACGGCTCA
TTCTTCCTGTACAGCGAGCTGACAGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC

TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
AGCCCCGGCAAG

21R75 Heavy chain nucleotide sequence with signal sequence (13B Version S1-2) (SEQ ID NO:61)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCGCTGGATATAAAGATGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC
TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA
TCTCCTGGCAAG

21R83 Heavy chain without predicted signal sequence (SEQ ID NO:62)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYISNYNRATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSSA
STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
LYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKCCVECPPCPAPPVAGPSVFL
FPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTFRV
VSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQPREPQVYTLPPSREEMTKNQ
VSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGSFFLYSELTVDKSRWQQGNV
FSCSVMHEALHNHYTQKSLSLSPGK

21R83 Heavy chain with predicted signal sequence (underlined) (SEQ ID NO:63)
MKHLWFFLLLVAAPRWVLSQVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAP
GQGLEWIGYISNYNRATNYNQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYD
YDVGMDYWGQGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWN
SGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSNFGTQTYTCNVDHKPSNTKVDKTVERKC
CVECPPCPAPPVAGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVQFNWYVDGVE
VHNAKTKPREEQFNSTFRVVSVLTVVHQDWLNGKEYKCKVSNKGLPAPIEKTISKTKGQP
REPQVYTLPPSREEMTKNQVSLTCLVEGFYPSDIAVEWESNGQPENNYKTTPPMLDSDGS
FFLYSELTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

21R83 Heavy chain variable region (SEQ ID NO:64)
QVQLVQSGAEVKKPGASVKISCKASGYSFTAYYIHWVKQAPGQGLEWIGYISNYNRATNY
NQKFKGRVTFTTDTSTSTAYMELRSLRSDDTAVYYCARDYDYDVGMDYWGQGTLVTVSS

21R83 Heavy chain CDR2 (SEQ ID NO:65)

YISNYNRATNYNQKFKG

21R83 Heavy chain nucleotide sequence with signal sequence underlined (13B Version 1) (SEQ ID NO:66)

ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCCCCT
GGACAGGGCCTGGAATGGATCGGCTACATCTCCAACTACAACCGGGCCACCAATTACAAC
CAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCTACCTCTACCGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC
TACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCTAGCGCTTCC
ACCAAGGGCCCCTCCGTGTTTCCTCTGGCCCCTTGCTCCAGATCCACCTCCGAGTCTACC
GCCGCTCTGGGCTGCCTCGTGAAGGACTACTTCCCCGAGCCCGTGACAGTGTCCTGGAAC
TCTGGCGCTCTGACCTCCGGCGTGCACACCTTTCCAGCTGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCCTCCGTCGTGACTGTGCCCTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCCTCCAACACCAAGGTGGACAAGACCGTGGAACGGAAGTGC
TGCGTGGAATGCCCCCCTTGTCCTGCCCCTCCTGTGGCTGGCCCTAGCGTGTTCCTGTTC
CCCCCAAAGCCCAAGGACACCCTGATGATCTCCCGGACCCCCGAAGTGACCTGCGTGGTG
GTGGATGTGTCCCACGAGGACCCCGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAA
GTGCACAACGCCAAGACCAAGCCCAGAGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCCGTGCTGACCGTGGTGCATCAGGACTGGCTGAACGGCAAAGAGTACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCCATCGAAAAGACCATCTCTAAGACCAAGGGACAGCCC
CGCGAGCCCCAGGTGTACACACTGCCTCCATCCCGGGAAGAGATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAAGGCTTCTACCCCTCCGATATCGCCGTGGAATGGGAGTCC
AACGGCCAGCCCGAGAACAACTACAAGACCACCCCCCCCATGCTGGACTCCGACGGCTCA
TTCTTCCTGTACAGCGAGCTGACAGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
AGCCCCGGCAAG

21R75 Heavy chain nucleotide sequence with signal sequence underlined (13B Version S1-2) (SEQ ID NO:67)

ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCCCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCGCTGGATATAAAGATGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC
TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA

TCTCCTGGCAAG

21R75 Heavy chain variable region nucleotide sequence (13B Version 1) (SEQ ID NO:68)
CAGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATC
TCCTGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCC
CCTGGACAGGGCCTGGAATGGATCGGCTATATCGCCGGCTACAAGGACGCCACCAACTAC
AACCAGAAATTCAAGGGCAGAGTGACCTTCACCACCGACACCTCCACCTCTACCGCCTAC
ATGGAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTAC
GACTACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCCTCT

21R75 Heavy chain variable region nucleotide sequence (13B Version 2) (SEQ ID NO:69)
CAGGTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATA
AGTTGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCA
CCAGGACAGGGACTTGAATGGATCGGATATATCGCTGGATATAAAGATGCTACAAACTAT
AACCAAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATAC
ATGGAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTAT
GATTATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCT

21R83 Heavy chain variable region nucleotide sequence (13B Version 1) (SEQ ID NO:70)
CAGGTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATC
TCCTGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCC
CCTGGACAGGGCCTGGAATGGATCGGCTACATCTCCAACTACAACCGGGCCACCAATTAC
AACCAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCTACCTCTACCGCCTAC
ATGGAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTAC
GACTACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCTAGC

21R75 Heavy chain variable region nucleotide sequence (13B Version 2) (SEQ ID NO:71)
CAGGTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATA
AGTTGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCA
CCAGGACAGGGACTTGAATGGATCGGATATATCGCTGGATATAAAGATGCTACAAACTAT
AACCAAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATAC
ATGGAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTAT
GATTATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCT

21R83 Heavy chain nucleotide sequence with signal sequence underlined (13B Version 2) (SEQ ID NO:72)
ATGAAGCACCTATGGTTCTTTCTATTATTAGTGGCCGCTCCCCGTTGGGTGTTATCGCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCTCCAATTATAATAGAGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC
TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT

CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA
TCTCCTGGCAAGTAG

21R83 Heavy chain variable region nucleotide sequence (13B Version 2) (SEQ ID NO:73)

CAGGTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATA
AGTTGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCA
CCAGGACAGGGACTTGAATGGATCGGATATATCTCCAATTATAATAGAGCTACAAACTAT
AACCAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATAC
ATGGAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTAT
GATTATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCT

21R75 Heavy chain nucleotide sequence with signal sequence underlined (13B Version 2) (SEQ ID NO:74)

ATGAAGCACCTATGGTTCTTTCTATTATTAGTGGCCGCTCCCCGTTGGGTGTTATCGCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCGCTGGATATAAAGATGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC
TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA
TCTCCTGGCAAGTAG

21M18 Heavy chain nucleotide sequence (version 2) (SEQ ID NO:75)

ATGAAGCACCTATGGTTCTTTCTATTATTAGTGGCCGCTCCCCGTTGGGTGTTATCGCAG
GTTCAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGT
TGCAAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCA
GGACAGGGACTTGAATGGATCGGATATATCTCCTCTTATAATGGAGCTACAAACTATAAC
CAAAAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATG
GAATTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGAT
TATGATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCTGCATCC
ACTAAGGGACCATCCGTGTTCCCTTTGGCCCCTTGCTCTCGTTCGACCTCTGAATCGACT
GCCGCTCTGGGATGCCTCGTGAAAGATTACTTCCCTGAGCCTGTGACCGTTTCCTGGAAC
TCGGGCGCCCTAACCTCTGGCGTGCACACATTCCCTGCCGTGCTACAGTCTTCTGGCCTA
TACTCTTTATCTTCGGTTGTTACCGTACCTTCTTCTAACTTCGGAACCCAAACTTACACC

TGTAACGTAGACCACAAGCCTTCGAACACCAAGGTGGACAAGACTGTTGAGCGAAAGTGC
TGCGTTGAGTGCCCTCCATGTCCTGCACCTCCTGTGGCTGGCCCTTCTGTGTTCCTGTTC
CCTCCAAAACCTAAGGACACTCTAATGATCTCTCGGACTCCTGAGGTGACTTGCGTGGTT
GTGGACGTGTCCCACGAGGACCCTGAGGTGCAGTTCAATTGGTACGTGGACGGAGTCGAG
GTGCACAATGCAAAGACCAAGCCTCGGGAGGAACAGTTCAACTCCACCTTCCGGGTGGTT
TCTGTGTTGACCGTTGTGCACCAAGACTGGCTGAACGGCAAAGAATACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCTATCGAAAAGACCATCAGCAAGACCAAGGGCCAGCCT
CGCGAGCCTCAGGTGTACACCCTGCCTCCCAGCCGGGAAGAAATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAGGGCTTCTACCCTTCCGACATCGCCGTTGAGTGGGAGTCT
AACGGACAGCCGGAGAACAACTACAAGACTACGCCTCCAATGCTGGACTCCGACGGCTCC
TTCTTCCTGTACTCCGAACTGACCGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCATGCTCCGTAATGCACGAAGCCTTGCACAATCACTACACTCAAAAGTCCCTATCCTTA
TCTCCTGGCAAGTAG

21M18 Heavy chain variable region (version 2) (SEQ ID NO:76)
CAGCTAGTTCAGTCTGGAGCGGAAGTTAAGAAACCTGGAGCATCCGTGAAAATAAGTTGC
AAGGCATCCGGTTACTCGTTCACCGCATACTATATCCACTGGGTTAAACAGGCACCAGGA
CAGGGACTTGAATGGATCGGATATATCTCCTCTTATAATGGAGCTACAAACTATAACCAA
AAATTCAAAGGACGCGTGACTTTCACAACTGACACCTCAACCTCGACAGCATACATGGAA
TTACGGTCCCTACGGTCTGACGACACTGCCGTTTACTATTGCGCTAGAGATTATGATTAT
GATGTTGGAATGGACTATTGGGGCCAGGGAACACTGGTGACAGTGTCTTCT

21R75 Heavy chain nucleotide sequence with signal sequence (13B Version 1T) (SEQ ID NO:77)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCTCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCCCCT
GGACAGGGCCTGGAATGGATCGGCTATATCGCCGGCTACAAGGACGCCACCAACTACAAC
CAGAAATTCAAGGGCAGAGTGACCTTCACCACCGACACCTCCACCTCTACCGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC
TACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCCTCTGCTTCC
ACCAAGGGCCCCTCCGTGTTTCCTCTGGCCCCTTGCTCCAGATCCACCTCCGAGTCTACC
GCCGCTCTGGGCTGCCTCGTGAAGGACTACTTCCCCGAGCCCGTGACAGTGTCTTGGAAC
TCTGGCGCCCTGACCTCCGGCGTGCACACCTTTCCAGCTGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCCTCCGTCGTGACTGTGCCCTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCCTCCAACACCAAGGTGGACAAGACCGTGGAACGGAAGTGC
TGCGTGGAATGCCCCCCTTGTCCTGCCCCTCCTGTGGCTGGCCCTAGCGTGTTCCTGTTC
CCCCCAAAGCCCAAGGACACCCTGATGATCTCCCGGACCCCCGAAGTGACCTGCGTGGTG
GTGGATGTGTCCCACGAGGACCCCGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAA
GTGCACAACGCCAAGACCAAGCCCAGAGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCCGTGCTGACCGTGGTGCATCAGGACTGGCTGAACGGCAAAGAGTACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCCATCGAAAAGACCATCTCTAAGACCAAGGGACAGCCC
CGCGAGCCCCAGGTGTACACACTGCCTCCATCCCGGGAAGAGATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAAGGCTTCTACCCCTCCGATATCGCCGTGGAATGGGAGTCC
AACGGCCAGCCCGAGAACAACTACAAGACCACCCCCCCCCATGCTGGACTCCGACGGCTCA
TTCTTCCTGTACAGCGAGCTGACAGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
AGCCCCGGCAAG

21R83 Heavy chain nucleotide sequence with signal sequence underlined (13B Version 1T) (SEQ ID NO:78)
ATGAAGCACCTGTGGTTCTTTCTGCTGCTGGTGGCCGCTCCCAGATGGGTGCTGTCTCAG
GTGCAGCTGGTGCAGTCTGGCGCCGAAGTGAAGAAACCTGGCGCCTCCGTGAAGATCTCC
TGCAAGGCCTCCGGCTACTCCTTCACCGCCTACTACATCCACTGGGTCAAGCAGGCCCCT
GGACAGGGCCTGGAATGGATCGGCTACATCTCCAACTACAACCGGGCCACCAATTACAAC

CAGAAATTCAAGGGCCGCGTGACCTTCACCACCGACACCTCTACCTCTACCGCCTACATG
GAACTGCGGTCCCTGCGGAGCGACGACACCGCCGTGTACTACTGCGCCAGAGACTACGAC
TACGACGTGGGCATGGACTACTGGGGCCAGGGCACACTCGTGACCGTGTCTAGCGCTTCC
ACCAAGGGCCCCTCCGTGTTTCCTCTGGCCCCTTGCTCCAGATCCACCTCCGAGTCTACC
GCCGCTCTGGGCTGCCTCGTGAAGGACTACTTCCCCGAGCCCGTGACAGTGTCCTGGAAC
TCTGGCGCTCTGACCTCCGGCGTGCACACCTTTCCAGCTGTGCTGCAGTCCTCCGGCCTG
TACTCCCTGTCCTCCGTCGTGACTGTGCCCTCCTCCAACTTCGGCACCCAGACCTACACC
TGTAACGTGGACCACAAGCCCTCCAACACCAAGGTGGACAAGACCGTGGAACGGAAGTGC
TGCGTGGAATGCCCCCCTTGTCCTGCCCCTCCTGTGGCTGGCCCTAGCGTGTTCCTGTTC
CCCCCAAAGCCCAAGGACACCCTGATGATCTCCCGGACCCCCGAAGTGACCTGCGTGGTG
GTGGATGTGTCCCACGAGGACCCCGAGGTGCAGTTCAATTGGTACGTGGACGGCGTGGAA
GTGCACAACGCCAAGACCAAGCCCAGAGAGGAACAGTTCAACTCCACCTTCCGGGTGGTG
TCCGTGCTGACCGTGGTGCATCAGGACTGGCTGAACGGCAAAGAGTACAAGTGCAAGGTG
TCCAACAAGGGCCTGCCTGCCCCCATCGAAAAGACCATCTCTAAGACCAAGGGACAGCCC
CGCGAGCCCCAGGTGTACACACTGCCTCCATCCCGGGAAGAGATGACCAAGAACCAGGTG
TCCCTGACCTGTCTGGTGGAAGGCTTCTACCCCTCCGATATCGCCGTGGAATGGGAGTCC
AACGGCCAGCCCGAGAACAACTACAAGACCACCCCCCCCATGCTGGACTCCGACGGCTCA
TTCTTCCTGTACAGCGAGCTGACAGTGGACAAGTCCCGGTGGCAGCAGGGCAACGTGTTC
TCCTGCTCCGTGATGCACGAGGCCCTGCACAACCACTACACCCAGAAGTCCCTGTCCCTG
AGCCCCGGCAAG

Alternative 21R75, 21R79, 21R83, and 21M18 Heavy chain CDR1 (SEQ ID NO:79)
AYYIH

Anti-DLL4 heavy chain CDR2 consensus sequence (SEQ ID NO:80)
$YIX_1X_2YX_3X_4ATNYNQKFKG$
where X_1 is serine or alanine, X_2 is serine, asparagine, or glycine, X_3 is asparagine or lysine, and X_4 is glycine, arginine, or aspartic acid

WHAT IS CLAIMED IS:

1. A method of inhibiting growth of a tumor comprising contacting the tumor or tumor cells with an effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

2. A method of inhibiting growth of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

3. A method of reducing the tumorigenicity of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

4. A method of reducing the frequency of cancer stem cells in a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

5. The method of any one of claims 1-4, wherein the tumor is a colorectal tumor.

6. A method of treating cancer in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

7. A method of modulating angiogenesis in a subject that has cancer comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with leucovorin, 5-fluorouracil, and irinotecan.

8. The method of claim 6 or 7, wherein the cancer is colorectal cancer.

9. The method of any one of claims 1-8, wherein the combination of the bispecific antibody and leucovorin, 5-fluorouracil, and irinotecan is used as a second-line treatment.

10. A method of inhibiting growth of an ovarian, primary peritoneal, or fallopian tumor that is platinum-resistant comprising contacting the tumor or tumor cells with an effective amount of a bispecific

antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

11. A method of inhibiting growth of an ovarian, primary peritoneal, or fallopian tumor that is platinum-resistant in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

12. A method of reducing the tumorigenicity of an ovarian, primary peritoneal, or fallopian tumor that is platinum-resistant in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

13. A method of reducing the frequency of cancer stem cells in an ovarian, primary peritoneal, or fallopian tumor that is platinum-resistant in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

14. A method of treating ovarian, primary peritoneal, or fallopian cancer that is platinum-resistant in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

15. A method of modulating angiogenesis in a subject that has ovarian, primary peritoneal, or fallopian cancer that is platinum-resistant comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

16. The method of any one of claims 10-15, wherein the bispecific antibody is administered in combination with paclitaxel.

17. The method of any of claims 10-16, wherein the bispecific antibody or combination including the bispecific antibody is administered following failure of more than two prior therapies and/or following prior administration of an anti-VEGF agent.

18. The method of claim 17, wherein the anti-VEGF agent is bevacizumab.

19. A method of inhibiting growth of a tumor comprising contacting the tumor or tumor cells with an effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and nab-paclitaxel (ABRAXANE®).

20. A method of inhibiting growth of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and ABRAXANE®.

21. A method of reducing the tumorigenicity of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and ABRAXANE®.

22. A method of reducing the frequency of cancer stem cells in a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and ABRAXANE®.

23. The method of any one of claims 19-22, wherein the tumor is a pancreatic tumor.

24. A method of treating cancer in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and ABRAXANE®.

25. A method of modulating angiogenesis in a subject that has cancer comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with gemcitabine and ABRAXANE®.

26. The method of claim 24 or 25, wherein the cancer is pancreatic cancer.

27. The method of any one of claims 19-26, wherein the combination of the bispecific antibody, gemticabine, and ABRAXANE® is used as a first-line treatment.

28. A method of inhibiting growth of a tumor comprising contacting the tumor or tumor cells with an effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

29. A method of inhibiting growth of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

30. A method of reducing the tumorigenicity of a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

31. A method of reducing the frequency of cancer stem cells in a tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

32. The method of any one of claims 28-31, wherein the tumor is an endometrial tumor.

33. A method of treating cancer in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

34. A method of modulating angiogenesis in a subject that has cancer comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with paclitaxel and carboplatin.

35. The method of claim 33 or 34, wherein the cancer is endometrial cancer.

36. The method of any one of claims 28-35, wherein the combination of the bispecific antibody, paclitaxel, and carboplatin is administered as a first-line treatment.

37. A method of inhibiting growth of an endometrial tumor comprising contacting the tumor or tumor cells with an effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

38. A method of inhibiting growth of an endometrial tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

39. A method of reducing the tumorigenicity of an endometrial tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

40. A method of reducing the frequency of cancer stem cells in an endometrial tumor in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

41. A method of treating endometrial cancer in a subject comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

42. A method of modulating angiogenesis in a subject that has endometrial cancer comprising administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

43. The method of any one of claims 37-42, wherein the bispecific antibody is administered in combination with paclitaxel and carboplatin.

44. The method of any of claims 37-43, wherein the bispecific antibody or combination including the bispecific antibody is administered as a first-line treatment.

45. The method of any one of claims 1-44, wherein:
(a) the first antigen-binding site comprises a heavy chain CDR1 comprising NYWMH (SEQ ID NO:17), a heavy chain CDR2 comprising DINPSNGRTSYKEKFKR (SEQ ID NO:18), and a heavy chain CDR3 comprising HYDDKYYPLMDY (SEQ ID NO:19);
(b) the second antigen-binding site comprises a heavy chain CDR1 comprising TAYYIH (SEQ ID NO:13), a heavy chain CDR2 comprising YISNYNRATNYNQKFKG (SEQ ID NO:65), and a heavy chain CDR3 comprising RDYDYDVGMDY (SEQ ID NO:16); and
(c) both the first and second antigen-binding sites comprise a light chain CDR1 comprising RASESVDNYGISFMK (SEQ ID NO:20), a light chain CDR2 comprising AASNQGS (SEQ ID NO:21), and a light chain CDR3 comprising QQSKEVPWTFGG (SEQ ID NO:22).

46. The method of any one of claims 1-45, wherein the bispecific antibody comprises:
(a) a first heavy chain variable region having at least about 90% sequence identity to SEQ ID NO:11;
(b) a second heavy chain variable region having at least about 90% sequence identity to SEQ ID NO:64; and
(c) a first and a second light chain variable region having at least about 90% sequence identity to SEQ ID NO:12.

47. The method of claim 46, wherein the bispecific antibody comprises:
(a) a first heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:11;
(b) a second heavy chain variable region having at least about 95% sequence identity to SEQ ID NO:64; and
(c) a first and a second light chain variable region having at least about 95% sequence identity to SEQ ID NO:12.

48. The method of claim 47, wherein the bispecific antibody comprises:
(a) a first heavy chain variable region of SEQ ID NO:11;
(b) a second heavy chain variable region of SEQ ID NO:64; and
(c) a first and a second light chain variable region of SEQ ID NO:12.

49. The method of any one of claims 1-48, wherein the bispecific antibody comprises a first CH3 domain and a second CH3 domain, each of which is modified to promote formation of heteromultimers.

50. The method of claim 49, wherein the first and second CH3 domains of the bispecific antibody are modified based upon electrostatic effects.

51. The method of any one of claims 1-50, wherein the bispecific antibody comprises a first human IgG2 constant region with amino acid substitutions at positions corresponding to positions 249 and 288 of SEQ ID NO:42, wherein the amino acids are replaced with glutamate or aspartate, and a second human IgG2 constant region with amino acid substitutions at positions corresponding to positions 236 and 278 of SEQ ID NO:42, wherein the amino acids are replaced with lysine.

52. The method of any one of claims 1-50, wherein the bispecific antibody comprises a first human IgG2 constant region with amino acid substitutions at positions corresponding to positions 236 and 278 of SEQ ID NO:42, wherein the amino acids are replaced with lysine, and a second human IgG2 constant region with amino acid substitutions at positions corresponding to positions 249 and 288 of SEQ ID NO:42, wherein the amino acids are replaced with glutamate or aspartate.

53. The method of any one of claims 1-50, wherein the bispecific antibody comprises:
(a) a first heavy chain of SEQ ID NO:7;
(b) a second heavy chain of SEQ ID NO:62; and
(c) a first and a second light chain of SEQ ID NO:8.

54. The method of any one of claims 1-44, wherein the bispecific antibody comprises:
(a) a first heavy chain variable region sequence that is the same as a polypeptide encoded by the plasmid deposited with ATCC having deposit no. PTA-13236;
(b) a second heavy chain variable region sequence that is the same as a polypeptide encoded by the plasmid deposited with ATCC having deposit no. PTA-13278; and

(c) a light chain variable region sequence that is the same as a polypeptide encoded by the plasmid deposited with ATCC having deposit no. PTA-13235.

55. The method of any one of claims 1-44, wherein the bispecific antibody comprises:
(a) a first heavy chain encoded by the plasmid deposited with ATCC having deposit no. PTA-13236;
(b) a second heavy chain encoded by the plasmid deposited with ATCC having deposit no. PTA-13278; and
(c) a light chain encoded by the plasmid deposited with ATCC having deposit no. PTA-13235.

56. The method of any one of claims 1-44 wherein the bispecific antibody is 219R45-MB-21R83 (305B83).

57. The method of any one of claims 1-56, wherein the tumor or cancer has metastasized.

58. The method of any one of claims 2-9, 11-18, 20-27, and 29-36, and 38-57, wherein the subject is human.

59. The method of any of claims 1-58, wherein the bispecific antibody is administered weekly, every other week, every three weeks, or every four weeks.

60. The method of claim 59, wherein the bispecific antibody is administered every two weeks or every three weeks.

61. The method of any of claims 1-60, wherein the bispecific antibody is administered at a dose of about 0.5 mg/kg to about 20 mg/kg.

62. The method of claim 61, wherein the bispecific antibody is administered at a dose of about 0.5 mg/kg, 1.0 mg/kg, 2.0 mg/kg, 3.0 mg/kg, 4.0 mg/kg, 5 mg/kg, or 10 mg/kg.

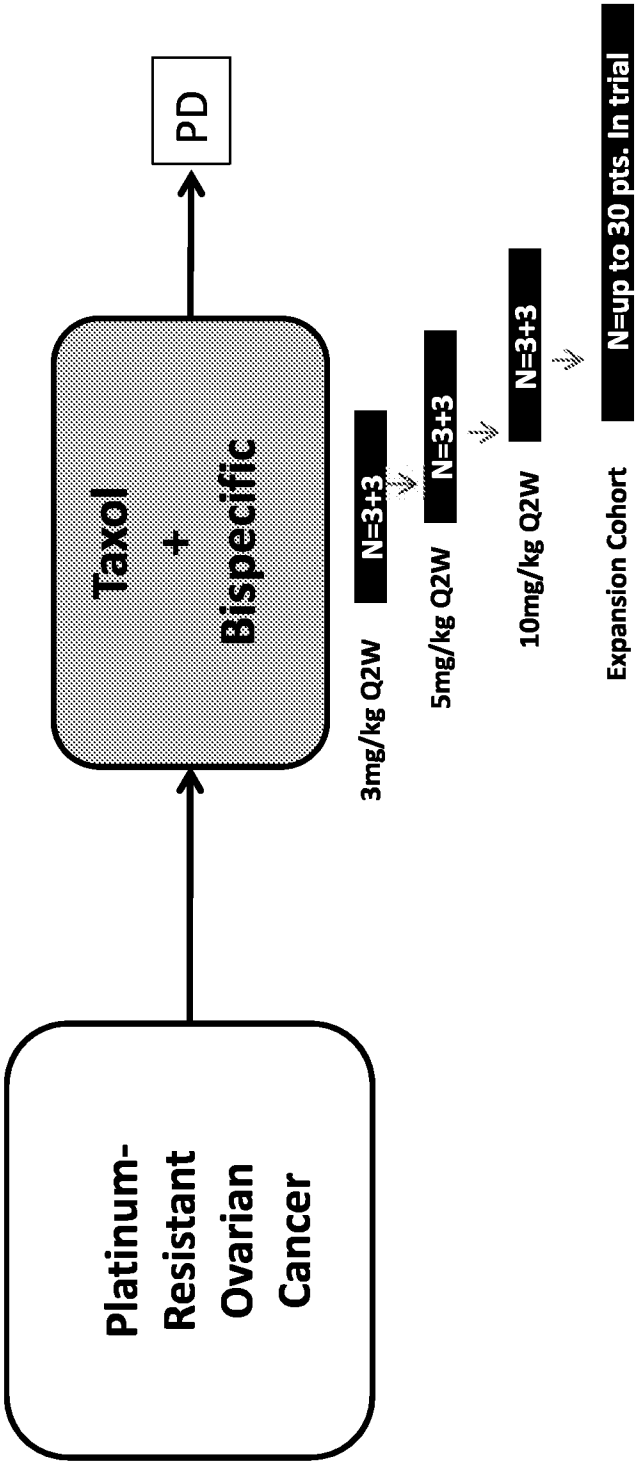
Platinum-Resistant Ovarian Cancer
Taxol + Bispecific
PD
3mg/kg Q2W
N=3+3
5mg/kg Q2W
N=3+3
10mg/kg Q2W
N=3+3
Expansion Cohort
N=up to 30 pts. In trial

Figure 1A

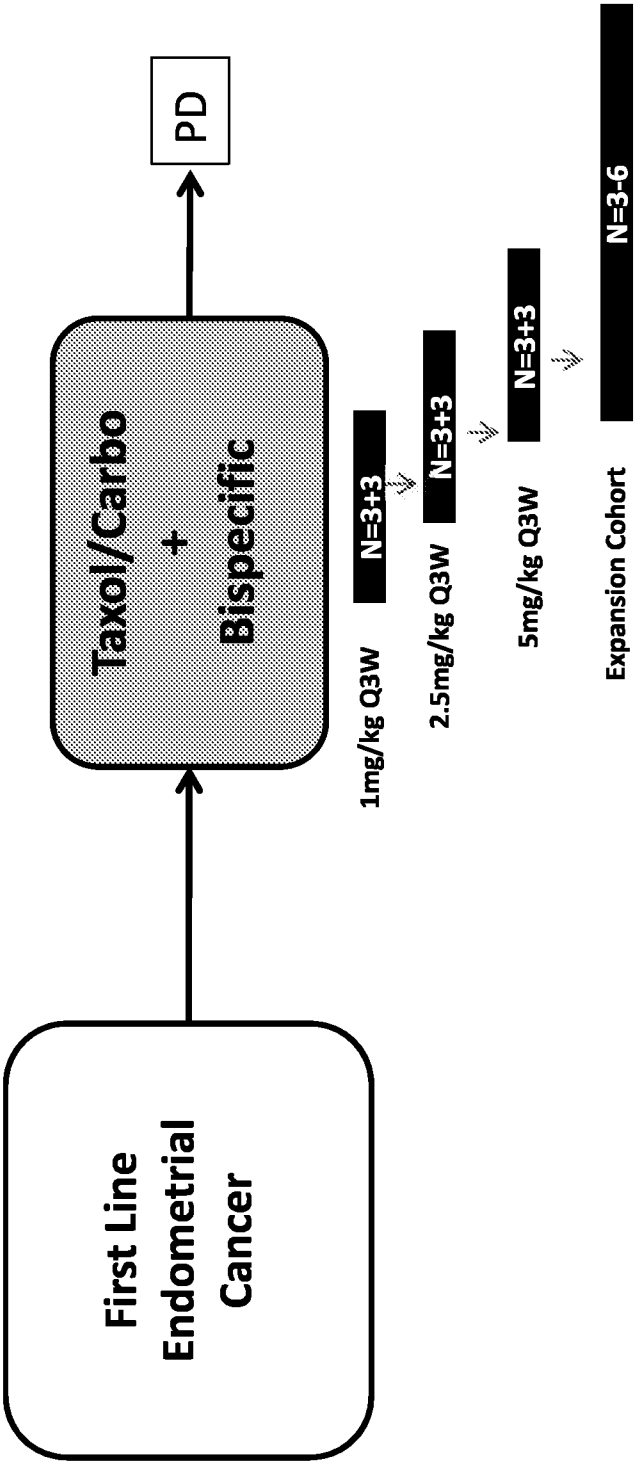
First Line Endometrial Cancer
Taxol/Carbo
+
Bispecific
PD
1mg/kg Q3W
N=3+3
2.5mg/kg Q3W
N=3+3
5mg/kg Q3W
N=3+3
Expansion Cohort
N=3-6

Figure 1B

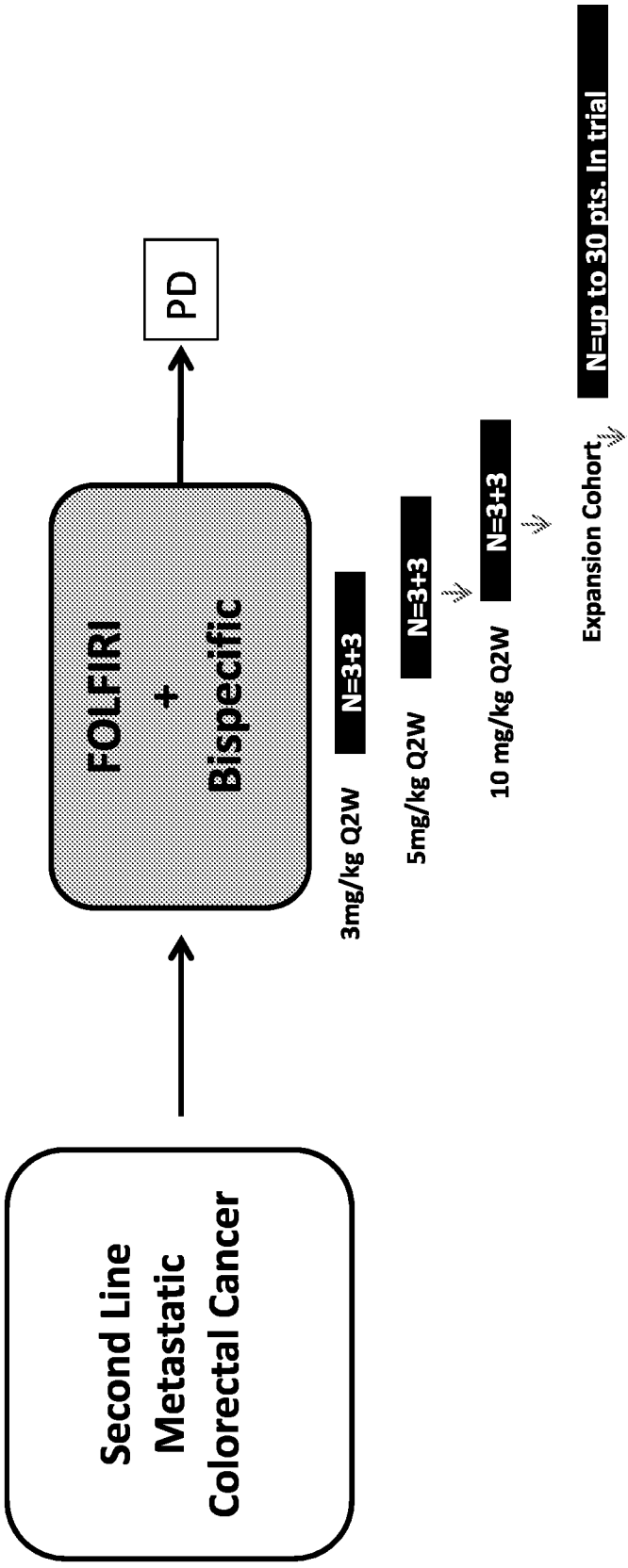
Second Line
Metastatic
Colorectal Cancer
FOLFIRI
+
Bispecific
PD
3mg/kg Q2W
N=3+3
5mg/kg Q2W
N=3+3
10 mg/kg Q2W
N=3+3
Expansion Cohort
N=up to 30 pts. In trial

Figure 2A

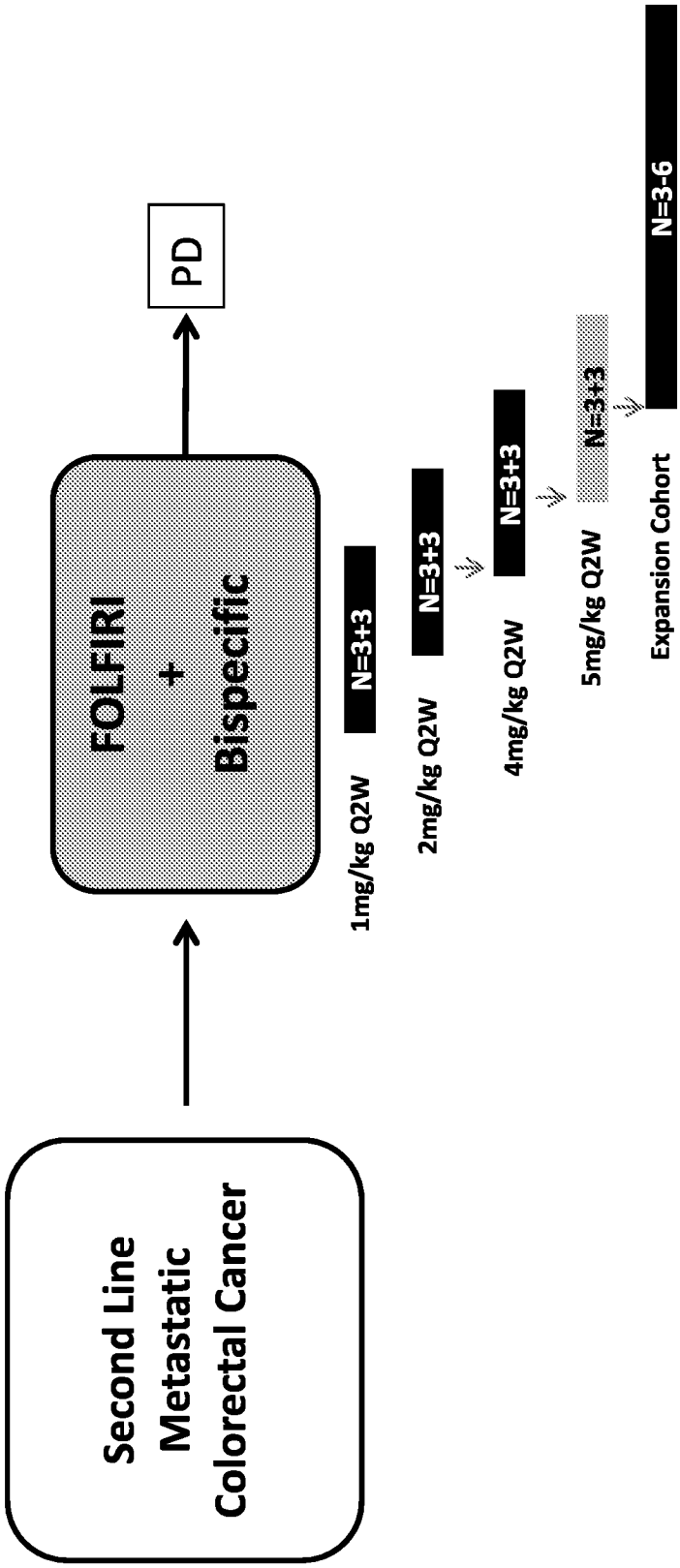
Second Line Metastatic Colorectal Cancer
FOLFIRI + Bispecific
PD
1mg/kg Q2W
N=3+3
2mg/kg Q2W
N=3+3
4mg/kg Q2W
N=3+3
5mg/kg Q2W
N=3+3
Expansion Cohort
N=3-6

Figure 2B

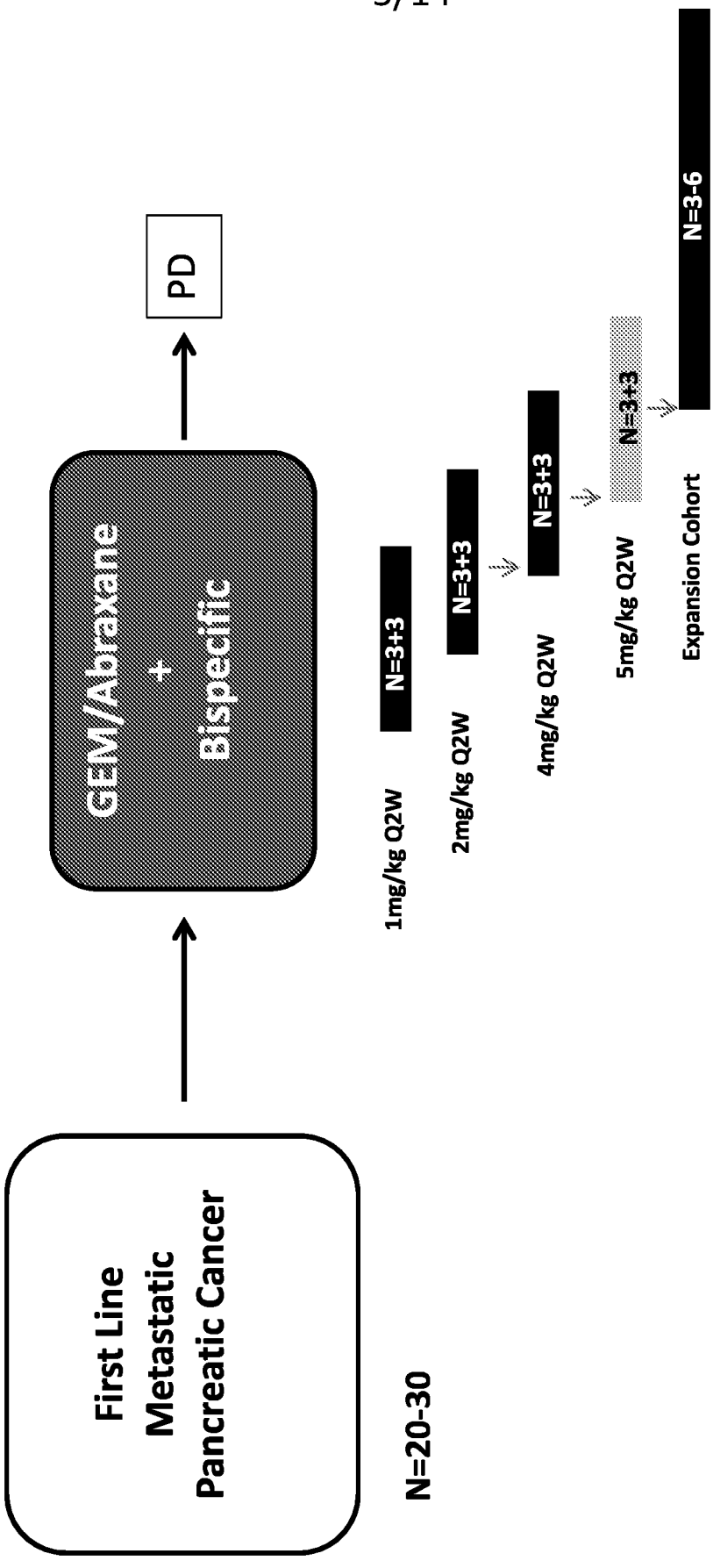
First Line
Metastatic
Pancreatic Cancer
N=20-30
GEM/Abraxane
+
Bispecific
PD
1mg/kg Q2W
N=3+3
2mg/kg Q2W
N=3+3
4mg/kg Q2W
N=3+3
5mg/kg Q2W
N=3+3
Expansion Cohort
N=3-6

Figure 3

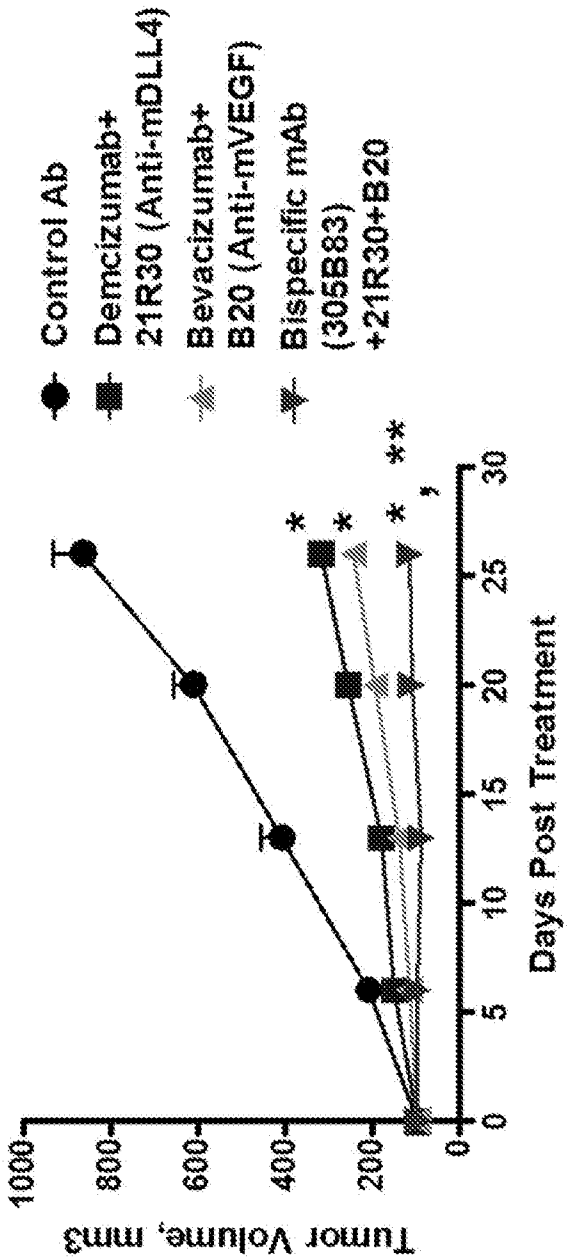
Control Ab
Demcizumab+
21R30 (Anti-mDLL4)
Bevacizumab+
B20 (Anti-mVEGF)
Bispecific mAb
(305B83)
+21R30+B20
Tumor Volume, mm3
1000
800
600
400
200
0
0
5
10
15
20
25
30
Days Post Treatment
*
*
*
**

Figure 4

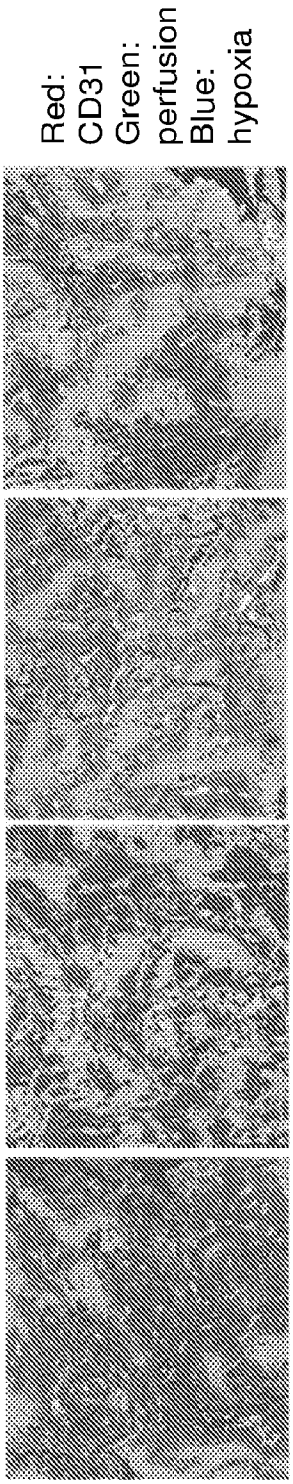
Red:
CD31
Green:
perfusion
Blue:
hypoxia
Dcontrol mAB
Demcizumab+
21R30 (Anti-mDLL4)
Bevacizumab+
B20 (Anti-mVEGF)
OMP-305B83+
21R30+B20

Figure 5A

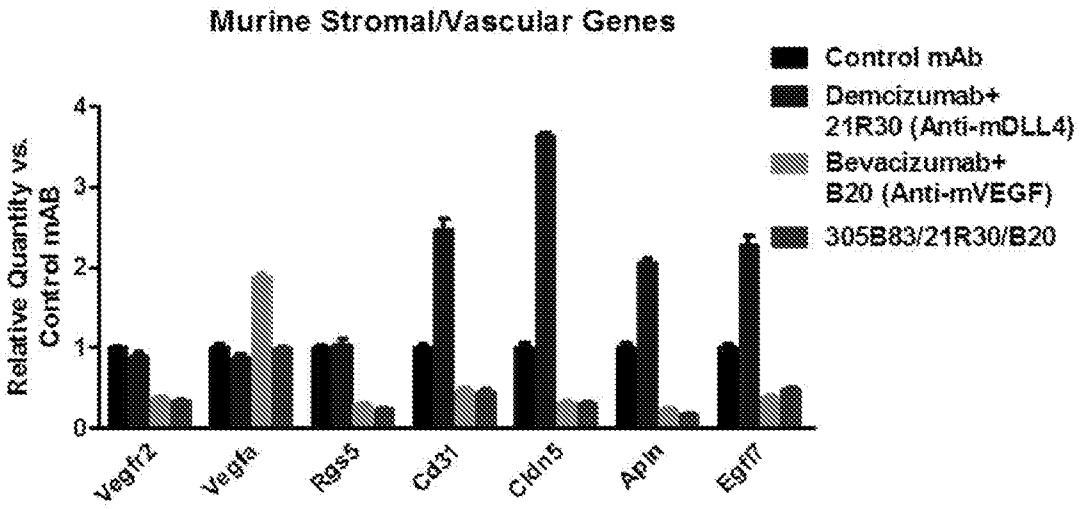

Figure 5B

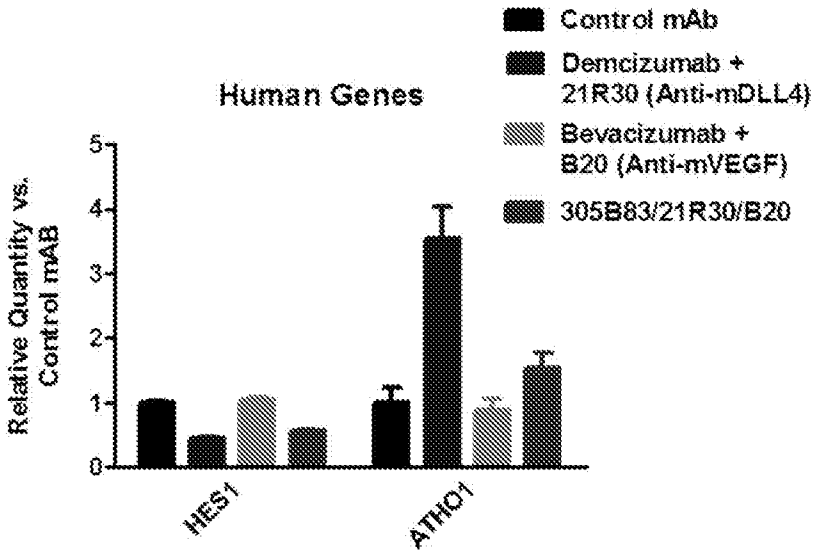

Figure 5C

OMP-OV40

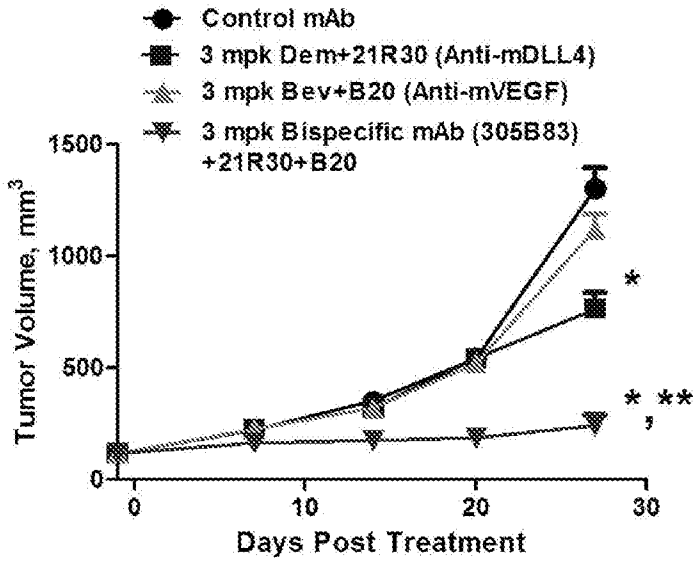

Figure 6A

OMP-STM1

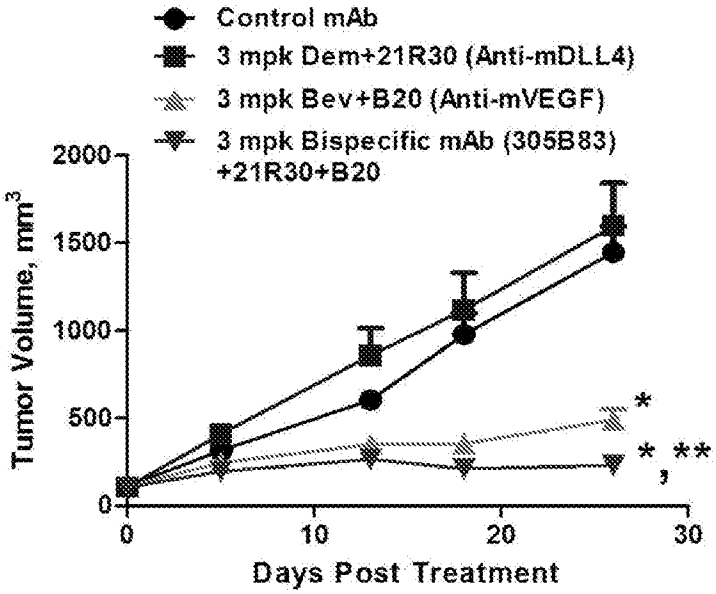

Figure 6B

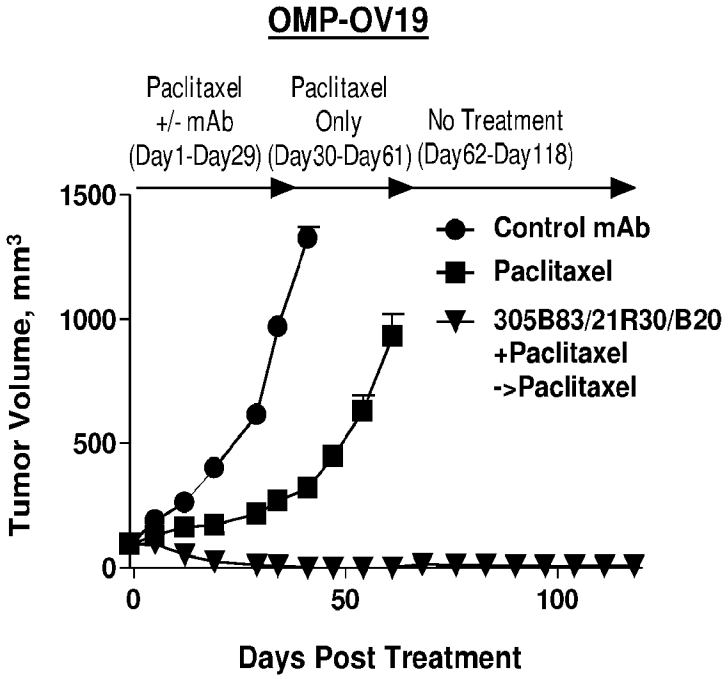

Figure 7A

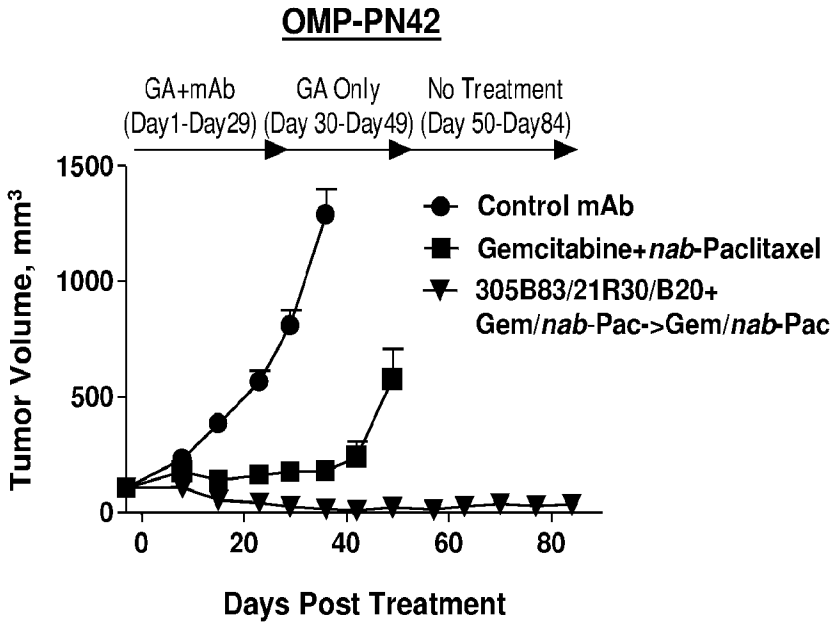

Figure 7B

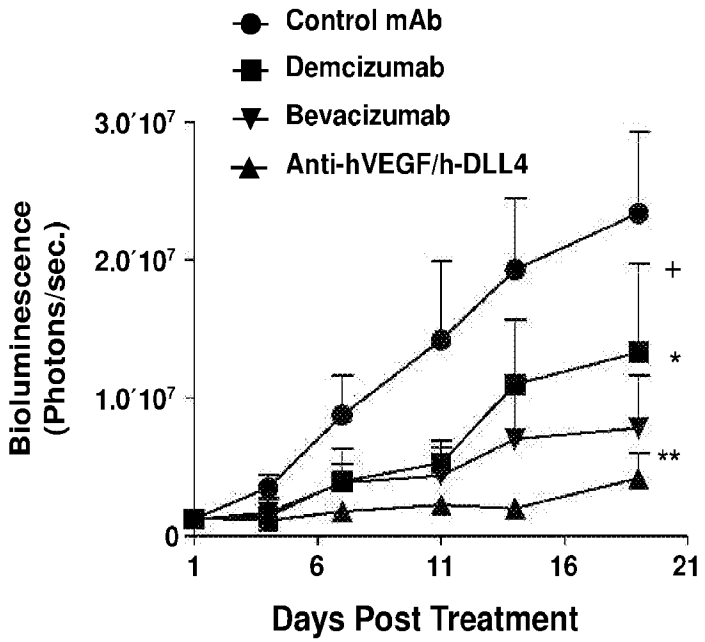

Figure 8A

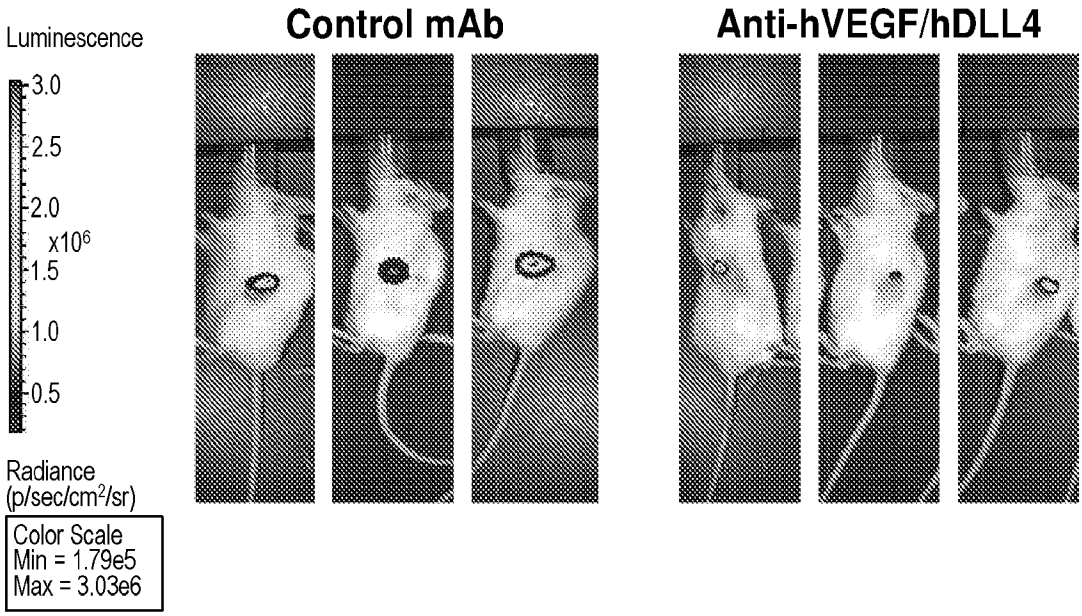

Figure 8B

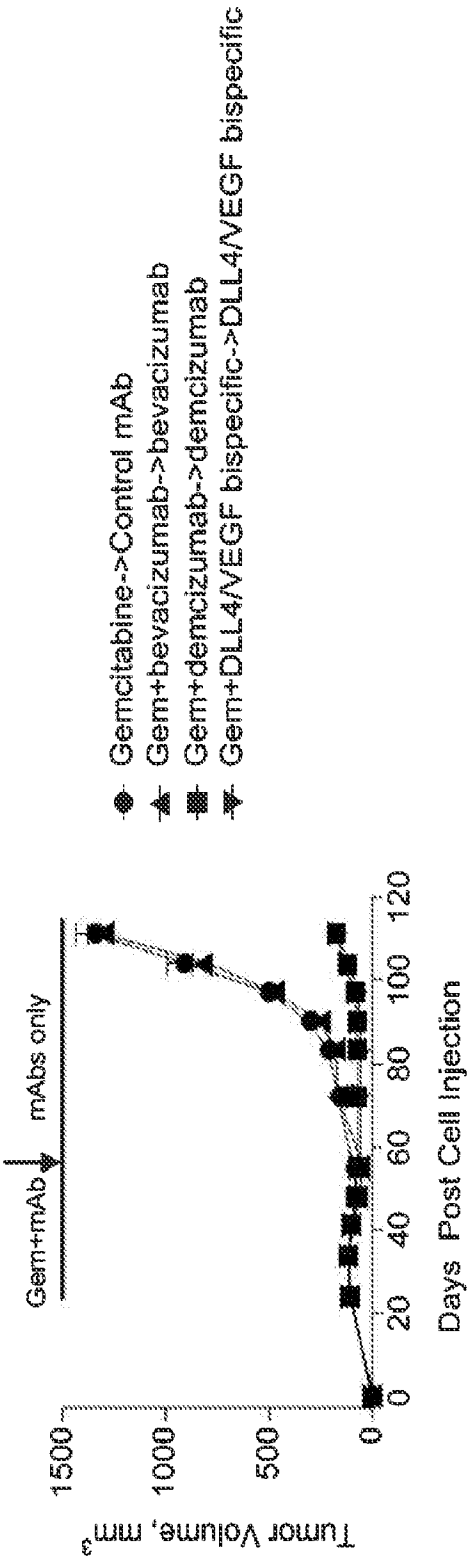
Gemcitabine->Control mAb
Gem+bevacizumab->bevacizumab
Gem+demcizumab->demcizumab
Gem+DLL4/VEGF bispecific->DLL4/VEGF bispecific
Gem+mAb
mAbs only
Tumor Volume, mm3
1500
1000
500
0
0
20
40
60
80
100
120
Days Post Cell Injection

Figure 9

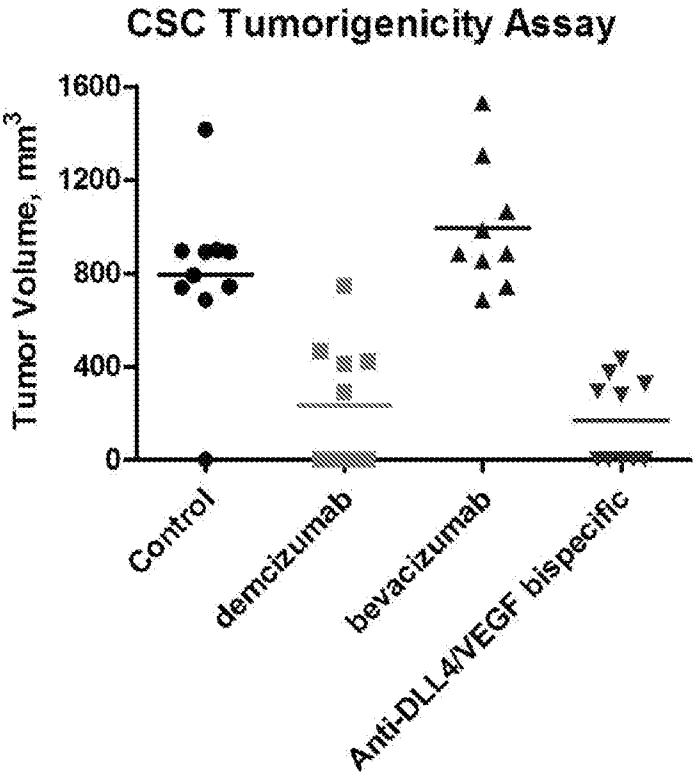
CSC Tumorigenicity Assay
Tumor Volume, mm3
1600
1200
800
400
0
Control
demcizumab
bevacizumab
Anti-DLL4/VEGF bispecific

Figure 10

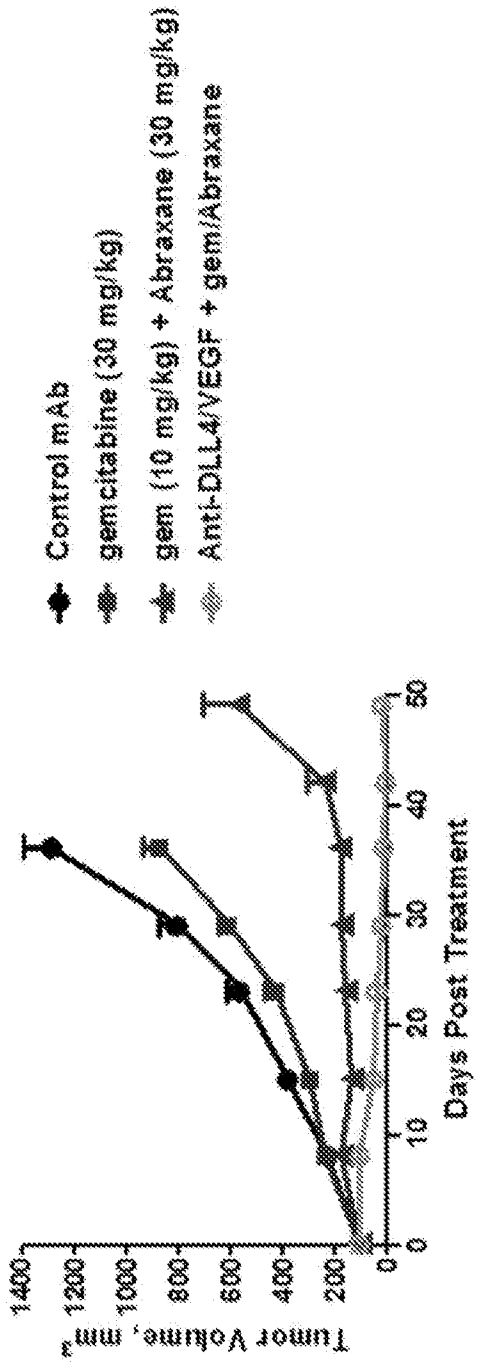
Control mAb
gemcitabine (30 mg/kg)
gem (10 mg/kg) + Abraxane (30 mg/kg)
Anti-DLL4/VEGF + gem/Abraxane
Tumor Volume, mm³
1400
1200
1000
800
600
400
200
0
10
20
30
40
50
Days Post Treatment

Figure 11

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/53316

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - C07K 16/22, 16/28, 16/46; A61K 31/282, 39/395 (2016.01)
CPC - C07K 16/22, 16/28, 16/468, 2317/31, 2317/35, 2317/56, 2317/76; A61K 2039/505, 2039/507, 39/395, 39/3955
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC(8): C07K 16/22, 16/28, 16/46; A61K 31/282, 39/395 (2016.01)
CPC: C07K 16/22, 16/28, 16/468, 2317/31, 2317/35, 2317/56, 2317/76; A61K 2039/505, 2039/507, 39/395, 39/3955

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
CPC: C07K 16/22, 16/28, 16/468, 2317/31, 2317/35, 2317/56, 2317/76; A61K 2039/505, 2039/507, 39/395, 39/3955 (text search)
USPC: 435/136.1; 530/387.3 (text search)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Electronic data bases: PatBase; Google Patents; Google Scholar Search terms: Bispecific antibody (BsAb), DLL4 [delta like ligand 4] and VEGF targets, Oncomed OMP-305B83 BsAb , targeting multiple signaling pathways, reducing tumorigenicity, reducing tumor growth, reducing frequency of cancer stem cells, modulating angiogenesis, chemotherape

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	US 2014/0348835 A1 (ABBVIE INC.) 27 November 2014 (27.11.2014). Especially para [0014], [0042], [0043], [0045]	1-3, 5/(1-3), 6-8, 37-39, 41, 42 --- 4, 5/4, 10-15, 40
Y	US 2008/0187532 A1 (Gurney et al.) 7 August 2008 (07.08.2008). Especially para [0033]	4, 5/4, 13, 40
Y	McAuliffe et al. Targeting Notch, a key pathway for ovarian cancer stem cells, sensitizes tumors to platinum therapy. Proc Nat Acad Sci 23 October 2012 Vol 109 Vol 43 Pages E2939-2849. Especially abstract.	10-15

☐ Further documents are listed in the continuation of Box C. ☐

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

Date of the actual completion of the international search	Date of mailing of the international search report
9 December 2016	21 FEB 2017
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300	Authorized officer: Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/53316

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

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

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

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

3. ☒ Claims Nos.: 9, 17, 18, 27, 36, 44-62
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

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

This International Searching Authority found multiple inventions in this international application, as follows:
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group I+: Claims 1-8, 10-16, 19-26, 28-35, 37-43, drawn to a method of treating a cancer in a subject comprising administrating a bispecific antibody (BsAb) comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4, in combination with chemotherapeutic agent(s).

-----Go to Extra Sheet for continuation-----

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

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

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

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
Claims 1-8, 10-15, 37-42 limited to BsAb (VEGF/DLL4) in combination with leucovorin, 5-fluorouracil, and irinotecan

Remark on Protest

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

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

☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/53316

------continuation of Box III (Lack of Unity of Invention)------

The method of treating a cancer will be searched to the extent that the combination therapy encompasses BsAb (VEGF/DLL4) in combination with leucovorin, 5-fluorouracil, and irinotecan (Claim 1). It is believed that claims 1-8, 10-15, 37-42 read on this first named invention and thus these claims will be searched without fee to the extent that they encompass BsAb (VEGF/DLL4) in combination with leucovorin, 5-fluorouracil, and irinotecan. Additional combination therapies will be searched upon payment of additional fees. Applicant must specify the claims that encompass any additional elected combination therapies Applicants must further indicate, if applicable, the claims which read on the first named invention if different than what was indicated above for this group. Failure to clearly identify how any paid additional invention fees are to be applied to the "+" group(s) will result in only the first claimed invention to be searched/examined. An exemplary election would be: BsAb (VEGF/DLL4) in combination with gemcitabine and nab-paclitaxel (claims 10 -16, 19-26, 37-42).

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

Special Technical Features:

Among the inventions listed as Groups I+ are cancer therapies comprising BsAb (VEGF/DLL4) in combination with chemotherapeutic agents, recited therein. Each invention requires a specific combination of BsAb (VEGF/DLL4) and chemotherapeutic agents, not required by any other inventions.

Common Technical Features:

1. Administering to the subject a therapeutically effective amount of a bispecific antibody comprising a first antigen-binding site that specifically binds human VEGF, and a second antigen-binding site that specifically binds human DLL4.

2. Treating a subject suffering from a specific type of cancer with the anti-VEGF, anti-DLL4 bispecific antibody in conjunction with chemotherapeutic agents.

3. A method of treating cancer, inhibiting growth of a tumor, reducing tumorigenicity, reducing the frequency of cancer stem cells, modulating angiogenesis.

However, said common technical feature does not represent a contribution over the prior art, and is obvious over the publication titled "Dual targeting of DLL4 and VEGF signaling by a novel bispecific antibody inhibits tumor growth and reduces cancer stem cell frequency" by YEN et al. (hereinafter "Yen")[available online 6 April 2014 as Abstract 207, AACR Annual Meeting. URL: http://www.abstractsonline.com/Plan/AbstractPrintView.aspx?mID=3404&sKey=df45d34f-08f7-4f5c-8fdc-4e9af3285b52&cKey=bdcc8883-9685-4dc4-b3cd-f9996ea8ceb8>], in view of US 2008/0187532 A1 to GURNEY et al. (hereinafter "Gurney")

As to common technical feature #1, Yen teaches (abstract; "We have recently developed a high binding affinity bispecific monoclonal antibody that targets both human DLL4 and human VEGF....The bispecific antibody demonstrated significant in vivo anti-tumor efficacy in various solid tumors, delayed tumor recurrence following termination of chemotherapy, and decreased the frequency of tumor initiating cell").

As to common technical feature #2, Gurney teaches (claim 36; An isolated antibody that specifically binds to a human DLL4 epitope comprising amino acids within human DLL4 N-terminal region (SEQ ID NO: 27), wherein the antibody inhibits DLL4 binding to a Notch receptor"; para [0131]; Exemplary bispecific antibodies can bind to two different epitopes, at least one of which originates in a polypeptide of the invention"; para [0181]; In other embodiments, the treatment can involve the combined administration of antibodies of the present invention with other antibodies against additional tumor associated antigens including, but not limited to, antibodies that bind....vascular endothelial growth factor (VEGF)"; para [0217-0218]; In Vivo Prevention and Treatment of Tumor Growth Using Anti-DLL4 Antibodies in Combination Therapy. DLL-4 Antibodies in Combination with Fluorouracil. [0218]; In certain embodiments, anti-DLL4 antibodies were analyzed in combination with chemotherapy for the ability to reduce growth of UM-C4 colon tumor cells").

As to common technical feature #3, Yen teaches treating cancer (abstract; "The bispecific antibody demonstrated significant in vivo anti-tumor efficacy in various solid tumors"), inhibiting growth of a tumor (abstract; "delayed tumor recurrence following termination of chemotherapy"), reducing tumorigenicity (abstract; "significant in vivo anti-tumor efficacy in various solid tumors"), reducing the frequency of cancer stem cells (abstract; "decreased the frequency of tumor initiating cells"), modulating angiogenesis (abstract; "inhibition of vascular gene expression and endothelial cell proliferation").

As the common technical features were known in the art at the time of the invention, they cannot be considered common special technical features that would otherwise unify the groups. The inventions lack unity with one another.

-----continued on next sheet-----

----continued from previous sheet----

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

Note concerning item 4: Claims 9, 17, 18, 27, 36, 44-62 are multiple dependent claims and are not drafted according to the second and third sentences of PCT Rule 6.4(a).

Note concerning claims 19-24. Claims 19-24 are objected to as indefinite. Claims 19-24 contain the trademark/trade name or commercial name "Abraxane". Where a trademark or trade name is used in a claim as a limitation to identify or describe a particular material or product, the claim does not comply with the requirements of PCT Rule 6.3(a). The claim scope is uncertain since the trademark or trade name cannot be used properly to identify any particular material or product. A trademark or trade name is used to identify a source of goods, and not the goods themselves. Thus, a trademark or trade name does not identify or describe the goods associated with the trademark or trade name. For the purposes of the International Search and Opinion, the compound name nab-paclitaxel will be used as a substitute for tradename Abraxane.