(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(72) Inventors: PORDY, Robert C; Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). SASIELA, William J; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). SCHWEMMER-GIPE, Daniel A.; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US).

(74) Agent: HEUSCH, Marina I. et al.; Swanson & Bratschun, L.L.C., 8210 Southpark Terrace, Colorado, Littleton, Colorado 80120 (US).

(54) Title: METHODS FOR TREATING PATIENTS WITH FAMILIAL HYPERCHOLESTEROLEMIA

(57) Abstract: The present invention provides methods for treating patients suffering from familial hypercholesterolemia, including both HeFH and HoFH. The methods of the invention provide for lowering at least one lipid parameter in the patient by administering a therapeutically effective amount of an antibody or antigen-binding fragment thereof that specifically binds to ANGPTL3 in combination with a therapeutically effective amount of a statin, a first lipid lowering agent other than a statin, and a second lipid lowering agent other than a statin. The first non-statin lipid lowering agent is an agent that inhibits cholesterol uptake (e.g. ezetimibe) and the second non-statin lipid-lowering agent is an inhibitor of microsomal triglyceride transfer protein (e.g. lomitapide). The combination therapy is useful in treating hypercholesterolemia, as well as hyperlipidemia, hyperlipoproteinemia and dyslipidemia, including hypertriglyceridemia, chylomicronemia, and to prevent or treat diseases or disorders, for which abnormal lipid metabolism is a risk factor, such as cardiovascular diseases.

METHODS FOR TREATING PATIENTS WITH FAMILIAL HYPERCHOLESTEROLEMIA

SEQUENCE STATEMENT

[0001] The instant application contains a Sequence Listing, which has been submitted electronically in ASCII format and is hereby incorporated by reference in its entirety. Said ASCII copy, created on April 25, 2017, is named SequenceList_26PCT.TXT and is 7 kilobytes in size.

FIELD OF THE INVENTION

[0002] The present invention relates to the field of therapeutic treatments of diseases and disorders, which are associated with elevated levels of lipids and lipoproteins. More specifically, the invention relates to the use of an ANGPTL3 inhibitor with concomitant lipid-lowering therapies to treat patients with familial hypercholesterolemia in order to achieve optimal serum lipid and lipoprotein levels.

BACKGROUND

[0003] Hyperlipidemia is a general term that encompasses diseases and disorders characterized by or associated with elevated levels of lipids and/or lipoproteins in the blood. Hyperlipidemias include hypercholesterolemia, hypertriglyceridemia, combined hyperlipidemia, and elevated lipoprotein a (Lp(a)). A particular prevalent form of hyperlipidemia in many populations is hypercholesterolemia.

[0004] Hypercholesterolemia, particularly an increase in low-density lipoprotein (LDL) cholesterol (LDL-C) levels, constitutes a major risk for the development of atherosclerosis and coronary heart disease (CHD) (Sharrett *et al.*, 2001, Circulation 104:1108-1113). Low-density lipoprotein cholesterol is identified as the primary target of cholesterol lowering therapy and is accepted as a valid surrogate therapeutic endpoint. Numerous studies have demonstrated that reducing LDL-C levels reduces the risk of CHD with a strong direct relationship between LDL-C levels and CHD events; for each 1 mmol/L (~40 mg/dL) reduction in LDL-C, cardiovascular disease (CVD) mortality and morbidity is lowered by 22%. Greater reductions in LDL-C produce greater reduction in events, and comparative data of intensive versus standard statin treatment suggest that the lower the LDL-C level, the greater the benefit in patients at very high cardiovascular (CV) risk.

[0005] Familial hypercholesterolemia (FH) is an inherited disorder of lipid metabolism that predisposes a person to premature severe cardiovascular disease (CVD) (Kolansky *et al.,* (2008), Am J Cardiology,102(11):1438-1443). FH can be either an autosomal dominant or an autosomal recessive disease that results from mutations in the low density lipoprotein receptor (LDLR), or in at least 3 different genes that code for proteins involved in hepatic clearance of

1

LDL-C can cause FH. Examples of such defects include mutations in the gene coding for the LDL receptor (LDLR) that removes LDL-C from the circulation, and in the gene for apolipoprotein (Apo) B, which is the major protein of the LDL particle. In all cases, FH is characterized by an accumulation of LDL-C in the plasma from birth and subsequent development of tendon xanthomas, xanthelasmas, atheromata, and CVD. FH can be classified as either heterozygous FH (heFH) or homozygous FH (hoFH) depending on whether the individual has a genetic defect in one (heterozygous) or both (homozygous) copies of the implicated gene.

[0006] Current LDL-C-lowering medications include statins, cholesterol absorption inhibitors, fibrates, niacin, bile acid sequestrants and Proprotein Convertase Subtilisin/Kexin Type 9 (PCSK9) inhibitors. Statins are a commonly prescribed treatment for LDL-C lowering. However, despite the availability of such lipid-lowering therapies, many high-risk patients fail to reach their guideline target LDL-C level (Gitt et al., 2010, Clin Res Cardiol 99(11):723-733). For patients who are still unable to achieve guideline target level for LDL-C, despite available lipid-modifying therapy (LMT), mechanical removal of LDL-C by lipoprotein apheresis (*e.g.*, LDL apheresis) is sometimes prescribed.

[0007] However, patients who are not at LDL-C goal despite receiving an optimized LMT regimen, would greatly benefit from alternative LDL-C lowering therapies, or through use of a combination of therapeutic agents, such as the agents and regimens described herein.

BRIEF SUMMARY OF THE INVENTION

[0007a] In its broadest aspect, the invention relates to methods of treating patients who suffer from familial hypercholesterolemia by administering an ANGPTL3 inhibitor in combination with other lipid modifying therapies to achieve optimal levels of serum lipids and lipoproteins.

[0007b] In a first aspect is provided an inhibitor of angiopoietin-like protein 3 (ANGPTL3) when used in a method of treating a patient suffering from familial hypercholesterolemia in combination with a statin and a lipid-lowering agent other than a statin, wherein the method comprises administering to the patient a therapeutically effective amount of the statin; the lipid lowering agent other than a statin; and inhibitor of ANGPTL3, wherein the antibody or antigen-binding fragment thereof that binds specifically to ANGTL3 comprises a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8; and the statin is selected from the group consisting of atorvastatin, pitavastatin, lovastatin, simvastatin, pravastatin, fluvastatin and rosuvastatin; and the lipid lowering agent other than a statin is an agent that inhibits cholesterol absorption, and wherein the patient has received or is receiving treatment with a lipid modifying therapy.

2

[0007c] In a second aspect is provided an ANGPTL3 inhibitor when used in a method for improving one or more lipid parameter(s) in a patient diagnosed with familial hypercholesterolemia, the method comprising administering one or more therapeutically effective doses of an ANGPTL3 inhibitor in combination with one or more therapeutically effective doses of a lipid lowering agent selected from the group consisting of a statin, an agent that inhibits cholesterol absorption, and an agent that inhibits microsomal triglyceride transfer protein (MTTP), or a combination thereof, wherein: the ANGPTL3 inhibitor is an antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 comprising a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8; the statin is selected from the group consisting of atorvastatin, pitavastatin, lovastatin, simvastatin, pravastatin, fluvastatin and rosuvastatin; the agent that inhibits cholesterol absorption is ezetimibe; and the agent that inhibits MTTP is lomitapide;

wherein the improvement in one or more lipid parameter(s) is one or more of the following:

(a) a decrease from baseline (week 0) in low density lipoprotein-C (LDL-C);

(b) a decrease from baseline in apolipoprotein B (Apo B);

(c) a decrease from baseline in non-high high density lipoprotein–C (non-HDL-C);

(d) a decrease from baseline in total cholesterol (total-C);

(e) a decrease from baseline lipoprotein (a) (Lp(a); and/or

(f) a decrease from baseline in triglycerides (TG);

and wherein the patient has received or is receiving treatment with a lipid modifying therapy.

[0008] In one embodiment, the method comprises administering to the patient suffering from familial hypercholesterolemia a therapeutically effective amount of a combination of (a) a statin; (b) one lipid lowering agent other than a statin and (c) an inhibitor of ANGPTL3.

[0009] In one embodiment, the patient is administered (a) a statin; (b) one lipid lowering agent other than a statin; (c) an inhibitor of ANGPTL3, and (d) a second lipid-lowering agent other than a statin.

[0010] In one embodiment, the familial hypercholesterolemia is selected from the group consisting of heterozygous familial hypercholesterolemia (HeFH) and homozygous familial hypercholesterolemia (HoFH).

[0011] In one embodiment, the statin is selected from the group consisting of atorvastatin (LIPITOR®), pitavastatin (LIVALO®), lovastatin (MEVACOR®), simvastatin (ZOCOR®), pravastatin (PRAVACHOL®) fluvastatin (LESCOL®) and rosuvastatin (CRESTOR®).

[0012] In one embodiment, the statin is rosuvastatin (CRESTOR®), which is administered orally once a day at a dose of about 5 mg to about 40 mg. In another embodiment, the statin is rosuvastatin (CRESTOR®), which is administered orally once a day at a dose of 5-40 mg.

[0013] In one embodiment, the statin is atorvastatin (LIPITOR®), which is administered orally once a day at a dose of about 10 mg to about 80 mg. In another embodiment, the statin is atorvastatin (LIPITOR®), which is administered orally once a day at a dose of 10-80 mg.

[0014] In one embodiment, the one lipid lowering agent other than a statin is an agent that inhibits cholesterol absorption.

[0015] In one embodiment, the agent that inhibits cholesterol absorption is ezetimibe (ZETIA®).

[0016] In one embodiment, the ezetimibe (ZETIA®) is administered orally once a day at a dose of about 10 mg. In another embodiment, the ezetimibe (ZETIA®) is administered orally once a day at a dose of 10 mg.

[0017] In one embodiment, the second lipid lowering agent other than a statin is an agent that inhibits microsomal triglyceride transfer protein (MTTP).

[0018] In one embodiment, the agent that inhibits microsomal triglyceride transfer protein is lomitapide (JUXTAPID®).

[0019] In one embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of about 5 mg to about 60 mg. In another embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of 5-60 mg.

[0020] In one embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of about 20 mg. In another embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of 20 mg.

[0021] In one embodiment, the second lipid lowering agent other than a statin is an agent that inhibits PCSK9. In one embodiment, the PCSK9 inhibitor is alirocumab (PRALUENT®).

[0022] In one embodiment, the second lipid lowering agent other than a statin is an agent that reduces the production of apoB-containing lipoproteins. In one embodiment, the agent that reduces the production of apoB containing lipoproteins is mipomersen.

[0023] It is also envisioned that additional agents that act to lower lipids may be substituted for the first and second lipid lowering agents described herein, or alternatively can be combined with the first and second lipid lowering agents, plus evinacumab to achieve normalization of at least one lipid parameter described herein.

[0024] In certain embodiments, the lipid lowering therapies described herein may be combined for use in treating patients undergoing apheresis, such that the level of one or more of the lipid parameters described herein is normalized.

[0025] In one embodiment, the ANGPTL3 inhibitor is selected from the group consisting of a small molecule inhibitor, a nucleic acid (e.g. an siRNA), and an antibody that binds specifically to ANGPTL3.

[0026] In one embodiment, the ANGPTL3 antibody is evinacumab.

[0027] In one embodiment, evinacumab is administered before, during, or after treatment with a statin, ezetimibe, lomitapide, mipomersen, a PCSK9 inhibitor, or any other lipid lowering agent established to be useful for achieving normalization of at least one lipid parameter described herein.

[0028] In one embodiment, evinacumab is administered intravenously at a dose ranging from about 1 mg/kg to about 20 mg/kg of body weight.

[0029] In one embodiment, evinacumab is administered intravenously at a dose of about 15 mg/kg of body weight. In another embodiment, evinacumab is administered intravenously at a dose of 15 mg/kg of body weight.

[0030] In one embodiment, evinacumab is administered subcutaneously at a dose ranging from about 50 mg to about 750 mg.

[0031] In one embodiment, evinacumab is administered subcutaneously at a dose ranging from about 250 mg to about 450 mg.

[0032] In one embodiment, evinacumab is administered every week, every two weeks, every 3 weeks, every 4 weeks, every 2 months, every 3 months, or every 4 months.

[0033] In a second aspect, the invention provides a method for improving one or more lipid parameter(s) in a patient diagnosed with familial hypercholesterolemia, the method comprising administering one or more therapeutically effective doses of an ANGPTL3 inhibitor in combination with one or more therapeutically effective doses of a lipid lowering agent selected from the group consisting of a statin, an agent that inhibits cholesterol absorption, an agent that inhibits microsomal triglyceride transfer protein (MTTP), or a combination thereof, wherein the improvement in one or more lipid parameter(s) is one or more of the following:

    (a)   a decrease from baseline (week 0) in low density lipoprotein-C (LDL-C);

    (b)   a decrease from baseline in apolipoprotein B (Apo B);

    (c)   a decrease from baseline in non-high high density lipoprotein–C (non-HDL-C);

    (d)   a decrease from baseline in total cholesterol (total-C);

    (e)   a decrease from baseline lipoprotein (a) (Lp(a); and/or

    (f)   a decrease from baseline in triglycerides (TG).

[0034] In one embodiment, the familial hypercholesterolemia is selected from the group consisting of heterozygous familial hypercholesterolemia (HeFH) and homozygous familial hypercholesterolemia (HoFH).

[0035] In one embodiment, the ANGPTL3 inhibitor is selected from the group consisting of a small molecule inhibitor, a nucleic acid (e.g. an siRNA), and an antibody that binds specifically to ANGPTL3.

[0036] In one embodiment, the antibody that binds specifically to ANGPTL3 is evinacumab.

[0037] In one embodiment, the statin is selected from the group consisting of atorvastatin (LIPITOR®), pitavastatin (LIVALO®), lovastatin (MEVACOR®), simvastatin (ZOCOR®), pravastatin (PRAVACHOL®) fluvastatin (LESCOL®) and rosuvastatin (CRESTOR®).

[0038] In one embodiment, the statin is rosuvastatin (CRESTOR®) and is administered orally once a day at a dose of about 5 mg to about 40 mg. In another embodiment, the statin is rosuvastatin (CRESTOR®) and is administered orally once a day at a dose of 5-40 mg.

[0039] In one embodiment, the statin is atorvastatin (LIPITOR®), and is administered orally once a day at a dose of about 10 mg to about 80 mg. In another embodiment, the statin is atorvastatin (LIPITOR®), and is administered orally once a day at a dose of 10-80 mg.

[0040] In one embodiment, the agent that inhibits cholesterol absorption is ezetimibe (ZETIA®).

[0041] In one embodiment, the ezetimibe (ZETIA®) is administered orally once a day at a dose of about 10 mg. In another embodiment, the ezetimibe (ZETIA®) is administered orally once a day at a dose of 10 mg.

[0042] In one embodiment, the agent that inhibits microsomal triglyceride transfer protein is lomitapide (JUXTAPID®).

[0043] In one embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of about 5 mg to about 60 mg. In another embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of 5-60 mg.

[0044] In one embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of about 20 mg. In another embodiment, the lomitapide (JUXTAPID®) is administered orally once a day at a dose of 20 mg.

[0045] In one embodiment, other lipid lowering agents may be combined with the agents noted above to achieve an acceptable level of at least one of the lipid parameters described above. Other agents include, but are not limited to PCSK9 inhibitors. In one embodiment, the PCSK9 inhibitor is an antibody that binds specifically to PCSK9. In one embodiment, the antibody that binds specifically to PCSK9 is alirocumab (PRALUENT®).

[0046] In one embodiment, an additional lipid lowering agent that can be combined with the therapies described above includes an agent that reduces the production of apoB-containing lipoproteins. In one embodiment, the agent that reduces the production of apoB containing lipoproteins is mipomersen.

[0047] It is also envisioned that additional agents that act to lower lipids may be substituted for the first and second lipid lowering agents described herein, or alternatively, can be combined with the first and second lipid lowering agents, plus evinacumab to achieve normal levels of at least one lipid parameter described herein.

[0048] In certain embodiments, the lipid lowering therapies described herein may be combined for use in treating patients undergoing apheresis, the goal being to lower the level of at least one or more of the lipid parameters described above to an acceptable range. In a related embodiment, the use of the combination of therapies described herein may eliminate the need for apheresis, or may help to increase the time interval between apheresis procedures.

[0049] In one embodiment, the treatment results in at least a 40% reduction from baseline in at least one lipid parameter.

[0050] In one embodiment, the treatment results in at least a 75% reduction from baseline in at least one lipid parameter.

[0051] In one embodiment, the treatment results in at least a 40% reduction from baseline in LDL-C levels.

[0052] In one embodiment, the antibody, or antigen-binding fragment thereof that binds specifically to ANGPTL3 comprises the complementary determining regions (CDRs) of a heavy chain variable (HCVR) having the amino acid sequence of SEQ ID NO: 1 and the CDRs of a light chain variable region (LCVR) of SEQ ID NO: 5.

[0053] In one embodiment, the antibody, or antigen-binding fragment thereof that binds specifically to ANGTL3 comprises a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8.

[0054] In one embodiment, the antibody, or antigen-binding fragment thereof that binds specifically to ANGPTL3 comprises a HCVR having the amino acid sequence of SEQ ID NO: 1 and a LCVR having the amino acid sequence of SEQ ID NO: 5.

[0055] In additional aspects, the invention provides uses of and pharmaceutical compositions comprising a combination of a statin, at least one lipid lowering agent other than a statin, and an inhibitor of angiopoietin-like protein 3 (ANGPTL3) in the treatment of a patient suffering from familial hypercholesterolemia. In still further aspects, the invention provides uses of and pharmaceutical compositions comprising an inhibitor of angiopoietin-like protein 3 (ANGPTL3) in combination with a lipid lowering agent selected from the group consisting of a statin, an agent that inhibits cholesterol absorption, and an agent that inhibits microsomal triglyceride transfer protein (MTTP), or a combination thereof, in the improvement of one or more lipid parameter(s) in a patient diagnosed with familial hypercholesterolemia.

[0056] Other embodiments of the present invention will become apparent from a review of the ensuing detailed description.

DETAILED DESCRIPTION

[0057] Before the present invention is described, it is to be understood that this invention is not limited to particular methods and experimental conditions described, as such methods and conditions may vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to be limiting, since the scope of the present invention will be limited only by the appended claims.

[0058] Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention

belongs. As used herein, the term "about," when used in reference to a particular recited numerical value, means that the value may vary from the recited value by no more than 1%. For example, as used herein, the expression "about 100" includes 99 and 101 and all values in between (*e.g.*, 99.1, 99.2, 99.3, 99.4, etc.).

[0059] Although any methods and materials similar or equivalent to those described herein can be used in the practice of the present invention, the preferred methods and materials are now described. All publications mentioned herein are incorporated herein by reference to describe in their entirety.

Methods for Treating Hyperlipidemias

[0060] The present invention relates generally to methods and compositions for reducing lipoprotein levels in patients suffering from familial hypercholesterolemia, by administering a combination of (a) a statin; (b) a first lipid lowering therapy other than a statin; and (c) an inhibitor of ANGPTL3. In certain embodiments, the combination includes a second lipid lowering agent other than a statin. In certain embodiments, the first lipid-lowering agent that is not a statin is an agent that inhibits cholesterol absorption, such as ezetimibe (ZETIA®). In certain embodiments, the second lipid lowering agent that is not a statin is an agent that inhibits microsomal triglyceride transfer protein, such as lomitapide (JUSTAPID®). In one embodiment, the ANGPTL3 inhibitor is an antibody that binds specifically to ANGPTL3, such as evinacumab. In certain embodiments of the invention, treatment with the combination of an ANGPTL3 inhibitor (e.g. evinacumab) with the other therapies noted above (a statin, ezetimibe and lomitapide), may serve to lower the levels of lipoproteins in these patients to an acceptable range, thereby lowering their risk for development of atherosclerosis, stroke and other cardiovascular diseases. In certain embodiments, the methods described may be used to treat patients suffering from familial hypercholesterolemia, including heterozygous familial hypercholesterolemia (HeFH) and/or homozygous familial hypercholesterolemia (HoFH). In certain embodiments, a PCSK9 inhibitor may also be added to the combined therapies described above to further lower the level of at least one lipid parameter described herein. In a related embodiment, the combination of therapies described above may also be used in patients that are undergoing apheresis to achieve normalization of at least one of the lipid parameters described. The combination of therapies described may eliminate the need for apheresis, or may increase the time interval between the need for apheresis procedures. The combination of therapies described, when used alone or in combination with apheresis may serve to lower the risk for the development of atherosclerosis and coronary heart disease (CHD) in these patients.

[0061] As used herein, the term "lipoprotein" means a biomolecular particle containing both protein and lipid. Examples of lipoproteins include, *e.g.*, low density lipoprotein (LDL), high-

density lipoprotein (HDL), very low density lipoprotein (VLDL), intermediate density lipoprotein (IDL), and lipoprotein (a) (Lp(a)).

[0062] The present invention, according to certain embodiments, includes methods for treating patients who are non-responsive to, inadequately controlled by, or intolerant to lipid modifying therapies, other than those described and included in the combination described herein. As used herein, a particular patient who is "non-responsive to, inadequately controlled by, or intolerant to, lipid modifying therapy" is determined by a physician, physician's assistant, diagnostician, or other medical professional on the basis of the level of one or more lipoproteins (e.g., LDL-C and/or non-HDL-C) measured or otherwise detected in the serum of the patient after treatment with the lipid modifying agent. The physician, physician's assistant, diagnostician, or other medical professional can also determine if the patient is intolerant to certain lipid modifying therapies based on the side effect profile of the lipid modifying therapies, which the patient may experience, including, but not limited to, muscle aches, tenderness or weakness (myalgia), headache, skin flushing, difficulty sleeping, abdominal cramping, bloating, diarrhea, constipation, rash, nausea, or vomiting. A patient who is non-responsive to, inadequately controlled by, or intolerant to certain lipid modifying therapy may also be determined or influenced by other factors such as the patient's family history, medical background, current therapeutic treatment status, as well as generally accepted or prevailing lipoprotein targets adopted by national medical associations and physicians' groups. For example, in certain contexts, if a patient is undergoing therapy with a certain lipid modifying agent, and exhibits an LDL-C level of greater than or equal to about 70 mg/dL, this indicates that the patient is "non-responsive to, or inadequately controlled by, or intolerant to that lipid modifying therapy" and may benefit by treatment using the therapies described herein. In other contexts, if a patient is undergoing therapy with a certain lipid modifying agent, and exhibits an LDL-C level of greater than or equal to about 100 mg/dL, this indicates that the patient is "non-responsive to, inadequately controlled by, or intolerant to that lipid modifying therapy" and may benefit by treatment using the therapies described herein. In certain contexts, if a patient is undergoing therapy with a certain lipid modifying agent, and exhibits an LDL-C level of greater than or equal to about 150 mg/dL, 200 mg/dL, 250 mg/dL, 300 mg/dL, 400 mg/dL or higher, this indicates that the patient is "non-responsive to, inadequately controlled by, or intolerant to a certain lipid modifying therapy" and may benefit by treatment using the therapies described herein. In yet other contexts, whether or not a particular percentage reduction in LDL-C or non-HDL-C level is met, relative to the patient's LDL-C or non-HDL-C level at a particular start point ("baseline") can be used to determine whether the patient has responded to a lipid modifying therapy or whether that patient is in need of further treatment using the methods and agents of the present invention. For instance, a reduction in LDL-C or non-HDL-C of less than 50% (e.g., less than 40%, less than 35%, less than 30%, less than 25%, etc.) from baseline may signify a need for therapy using the methods and agents of the invention.

8

[0063] The present invention, accordingly, includes methods of treatment comprising administering one or more doses of an ANGPTL3 inhibitor (e.g. evinacumab) and one or more doses of a combination of a statin, ezetimibe, a PCSK9 inhibitor, mipomersen and/or lomitapide to a patient who is undergoing other types of lipid modifying therapy (e.g. bile acid sequestrants, niacin, fenofibrate), but is non-responsive to such therapy, or is intolerant to such therapy, wherein, after receiving one or more doses of the combination therapy described herein, the patient is able to achieve normal levels of total cholesterol, LDL-C, or non-HDL-C. In certain instances, the patient may be taken off of the other lipid modifying therapy, or the other lipid modifying therapy may be continued, but may be administered at lower doses and may be used in combination with the ANGPTL3 inhibitor and a statin plus ezetimibe and lomitapide, and optionally, a PCSK9 inhibitor, and/or mipomersen to achieve and/or maintain a particular target lipoprotein level. Alternatively, the patient may be administered the other lipid modifying therapy at the normal prescribed dose, but the frequency of administration of the other lipid modifying therapy may be reduced if the other lipid modifying therapy is to be administered in conjunction with the combination described herein. In some instances, the need for treatment with the other lipid modifying therapy by the patient to achieve and/or maintain a particular target lipoprotein level may be eliminated altogether following administration of one or more doses of the combination therapies described herein.

[0064] According to certain embodiments, the present invention comprises methods for reducing or eliminating the need for certain lipid modifying therapy, wherein the methods comprise selecting a patient with hyperlipidemia (*e.g.*, hypercholesterolemia) who has been treated with certain lipid modifying therapies within the last month, the last 2 months, the last 3 months, the last 4 months, the last 5 months, the last 6 months, or for a longer period, and administering one or more doses of an ANGPTL3 inhibitor in combination with the agents described herein (ezetimibe, lomitapide and a statin) to the patient. The methods according to this aspect of the invention result in lowering the level of at least one lipid, or lipoprotein in the serum of the patient, and consequently allow for a reduction or elimination of the need for treatment with the other lipid modifying therapy to which the patient did not respond (e.g. a bile acid sequestrant, niacin, or fenofibrate), or for which the patient showed an intolerance. The methods described herein may also be used in patients undergoing apheresis and the combination of lipid lowering agents used in this patient population may result in elimination of the need for apheresis, or may increase the time interval between apheresis procedures. For example, in certain embodiments of the present invention, following administration of one or more doses of an ANGPLT3 inhibitor in combination with a statin, ezetimibe and/or lomitapide, the serum LDL-C level of the patient is reduced to less than a defined level (*e.g.*, less than 100 mg/dL or less than 70 mg/dL), or the total cholesterol is lowered to a defined level (*e.g.* less than 200 mg/dL, or less than 150 mg/dL, or the serum level of LDL-C shows at least a 40%

reduction compared to the baseline levels before treatment with the combination described herein.

[0065] According to certain embodiments, the patient who is treatable by the methods of the present invention has hypercholesterolemia (*e.g.*, a serum LDL-C concentration of greater than or equal to 70 mg/dL (*e.g.* if the patient has a history of a cardiovascular event), or a serum LDL-C concentration greater than or equal to 100 mg/dL (*e.g.* if the patient has no history of a cardiovascular event). In certain embodiments, the patient's hypercholesterolemia is inadequately controlled by certain standard lipid modifying therapies, such as bile acid sequestrants, niacin, or fenofibrates. The present invention also includes methods for reducing total cholesterol, LDL-C, non-HDL-C, triglycerides (TG), ApoB, ApoCIII and Lp(a) in a patient who has familial hypercholesterolemia, including HeFH and HoFH.

Patients Suitable for Treatment

[0066] The present invention includes methods and compositions useful for treating patients who are diagnosed with or identified as being at risk of developing a hypercholesterolemia condition such as, *e.g.*, Heterozygous Familial Hypercholesterolemia (HeFH) or Homozygous Familial Hypercholesterolemia (HoFH) resulting from mutations in the low-density lipoprotein receptor (LDLR), Autosomal Dominant Hypercholesterolemia (ADH, *e.g.*, ADH associated with one or more gain-of-function mutations in the PCSK9 gene), documented presence of homozygous or compound heterozygous mutations in the Apo B gene, autosomal recessive hypercholesterolemia (ARH, *e.g.*, ARH associated with mutations in LDLRAP1), as well as incidences of hypercholesterolemia that are distinct from Familial Hypercholesterolemia (non FH). A patient who is suitable for treatment using the methods of the invention may also include patients who exhibit LDLR mutations that fall within any of the following classes: Class I: Receptor null mutations, whereby LDLR is not synthesized at all; Class II: Transport defective alleles, whereby LDLR is not properly transported from the endoplasmic reticulum to the Golgi apparatus for expression on the cell surface (class IIA (no receptor transport) and class IIB (reduced receptor transport); Class III: Binding defective alleles, whereby LDLR does not properly bind LDL on the cell surface because of a defect in either apolipoprotein B100 (R3500Q) or in LDL-R; Class IV: Internalization defective alleles whereby LDLR bound to LDL does not properly cluster in clathrin-coated pits for receptor-mediated endocytosis; Class V: Recycling defective alleles, whereby LDLR is not recycled back to the cell surface.

[0067] Diagnosis of familial hypercholesterolemia (*e.g.*, heFH or hoFH) can be made by genotyping and/or clinical criteria. For patients who are not genotyped, clinical diagnosis may be based on either the Simon Broome criteria with a criteria for definite FH, or the WHO/Dutch Lipid Network criteria with a score > 8 points.

[0068] According to certain embodiments, a patient may be suitable for treatment on the basis of having a history of coronary heart disease (CHD). As used herein a "history of CHD" (or

"documented history of CHD") includes one or more of: (i) acute myocardial infarction (MI); (ii) silent MI; (iii) unstable angina; (iv) coronary revascularization procedure (e.g., percutaneous coronary intervention [PCI] or coronary artery bypass graft surgery [CABG]); and/or (v) clinically significant CHD diagnosed by invasive or non-invasive testing (such as coronary angiography, stress test using treadmill, stress echocardiography or nuclear imaging).

[0069] According to certain embodiments, a patient may be suitable for treatment on the basis of having non-coronary heart disease cardiovascular disease ("non-CHD CVD"). As used herein, "non-CHD CVD" includes one or more of: (i) documented previous ischemic stroke with a focal ischemic neurological deficit that persisted more than 24 hours, considered as being of atherothrombotic origin; (ii) peripheral arterial disease; (iii) abdominal aortic aneurysm; (iv) atherosclerotic renal artery stenosis; and/or (v) carotid artery disease (transient ischemic attacks or >50% obstruction of a carotid artery).

[0070] According to certain embodiments, a patient may be suitable for treatment on the basis of having one or more additional risk factors such as, e.g., (i) documented moderate chronic kidney disease (CKD) as defined by 30 $\leq$eGFR <60 mL/min/1.73 m2 for 3 months or more; (ii) type 1 or type 2 diabetes mellitus with or without target organ damage (e.g., retinopathy, nephropathy, microalbuminuria); (iii) a calculated 10-year fatal CVD risk SCORE $\geq$5% (ESC/EAS Guidelines for the management of dyslipidemias, Conroy et al., 2003, Eur. Heart J. 24:987-1003).

[0071] According to certain embodiments, a patient may be suitable for treatment on the basis of having one or more additional risk factors selected from the group consisting of age (e.g., older than 40, 45, 50, 55, 60, 65, 70, 75, or 80 years), race, national origin, gender (male or female), exercise habits (e.g., regular exerciser, non-exerciser), other preexisting medical conditions (e.g., type-II diabetes, high blood pressure, etc.), and current medication status (e.g., currently taking beta blockers, niacin, ezetimibe, fibrates, omega-3 fatty acids, bile acid resins, etc.).

[0072] According to certain embodiments of the present invention, a subject who is treatable by the methods of the invention may exhibit an elevated level of one or more inflammatory marker. Any marker of systemic inflammation can be utilized for the purposes of the present invention. Suitable inflammatory markers include, without limitation, C-reactive protein, cytokines (e.g., Il-6, IL-8, and/or IL-17), and cellular adhesion molecules (e.g., ICAM-1, ICAM-3, BL-CAM, LFA-2, VCAM-1, NCAM, and PECAM).

[0073] According to the present invention, patients may be suitable for treatment on the basis of a combination of one or more of the foregoing criteria or therapeutic characteristics. For example, according to certain embodiments, a patient suitable for treatment with the methods of the present invention, may further be selected on the basis of having HeFH or non-FH in combination with: (i) a history of documented CHD, (ii) non-CHD CVD, and/or (iii) diabetes

mellitus with target organ damage; such patients may also be selected on the basis of having a serum LDL-C concentration of greater than or equal to 70 mg/dL.

[0074] According to certain other embodiments, a patient suitable for treatment with the methods of the present invention, in addition to having hypercholesterolemia that is not adequately controlled by a daily moderate-dose therapeutic statin regimen, may further be selected on the basis of having HeFH or non-FH without CHD, or non-CHD CVD, but having either (i) a calculated 10-year fatal CVD risk SCORE ≥5%; or (ii) diabetes mellitus without target organ damage; such patients may also be selected on the basis of having a serum LDL-C concentration of greater than or equal to 100 mg/dL.

[0075] According to certain embodiments of the present invention, the subject who is treatable by the methods of the invention is a subject who has familial chylomicronemia syndrome (FCS; also known as lipoprotein lipase deficiency).

[0076] According to certain embodiments of the present invention, the subject who is treatable by the methods of the invention is a subject who is undergoing, or has recently undergone, lipoprotein apheresis (*e.g.*, within the last six months, within the last 12 weeks, within the last 8 weeks, within the last 6 weeks, within the last 4 weeks, within the last 2 weeks, etc.).

Administration of an ANGPTL3 Inhibitor as Add-On Therapy

[0077] The present invention includes methods of treatment wherein a patient who is undergoing, or has recently undergone, standard lipid modifying therapy (*e.g.* a statin) is administered an ANGPTL3 inhibitor according to a particular dosing amount and frequency, and wherein the ANGPTL3 inhibitor is administered as an add-on to the patient's pre-existing lipid modifying therapy (if applicable), such as an add-on to the patient's pre-existing daily therapeutic statin regimen, or other regimen, e.g. niacin. The methods also include use of the ANGPTL3 inhibitor (e.g. evinacumab) as add on therapy with lipid modifying therapies in addition to statins, including use with ezetimibe and lomitapide to achieve maximal lipid lowering effects. Additional lipid lowering agents to be used in the methods of the invention include PCSK9 inhibitors, or mipomersen. The combination of agents may also be used in patients undergoing apheresis to achieve acceptable lipid levels.

[0078] For example, the methods of the present invention include add-on therapeutic regimens wherein the ANGPTL3 inhibitor is administered as add-on therapy to the same stable daily therapeutic statin regimen (*i.e.*, same dosing amount of statin) that the patient was on prior to receiving the ANGPTL3 inhibitor. In addition to the statin plus the ANGPTL3 antibody therapy, the addition of either ezetimibe alone, or in combination with lomitapide results in significantly lower levels of serum lipids or lipoproteins when the combination is administered. In other embodiments, the ANGPTL3 inhibitors are administered as add-on therapy to a therapeutic statin regimen comprising a statin in an amount that is more than or less than the dose of statin the patient was on prior to receiving the ANGPTL3 inhibitor, or the combination therapy

described herein. For example, after starting a therapeutic regimen comprising an ANGPTL3 inhibitor administered at particular dosing frequencies and amounts, in addition to ezetimibe and lomitapide, the daily dose of statin administered or prescribed to the patient may (a) stay the same, (b) increase, or (c) decrease (*e.g.*, up-titrate or down-titrate) in comparison to the daily statin dose the patient was taking before starting the ANGPTL3 inhibitor, ezetimibe and/or lomitapide therapeutic regimen, depending on the therapeutic needs of the patient.

Therapeutic Efficacy

[0079] The methods of the present invention may result in the reduction in serum levels of one or more lipid components selected from the group consisting of total cholesterol, LDL-C, IDL, non-HDL-C, ApoB 100, ApoB 48, Apo A-1, Apo CIII, VLDL-C, triglycerides, Lp(a), chylomicrons, chylomicron remnants, and remnant cholesterol. For example, according to certain embodiments of the present invention, administration of an ANGPTL3 inhibitor in combination with a statin, ezetimibe and/or lomitapide to a suitable subject will result in a mean percent reduction from baseline in serum low density lipoprotein cholesterol (LDL-C) of at least about 25%, 30%, 40%, 50%, 60%, or greater; a mean percent reduction from baseline in ApoB of at least about 25%, 30%, 40%, 50%, 60%, or greater; a mean percent reduction from baseline in non-HDL-C of at least about 25%, 30%, 40%, 50%, 60%, or greater; a mean percent reduction from baseline in total cholesterol of at least about 10%, 15%, 20%, 25%, 30%, 35%, or greater; a mean percent reduction from baseline in VLDL-C of at least about 5%, 10%, 15%, 20%, 25%, 30%, or greater; a mean percent reduction from baseline in triglycerides of at least about 5%, 10%, 15%, 20%, 25%, 30%, 35% or greater; and/or a mean percent reduction from baseline in Lp(a) of at least about 5%, 10%, 15%, 20%, 25%, or greater.

ANGPTL3 Inhibitors

[0080] The methods of the present invention comprise administering to a patient a therapeutic composition comprising an ANGPTL3 inhibitor (e.g. an ANGPTL3 antibody such as evinacumab) in combination with a statin, an inhibitor of cholesterol absorption (e.g. ezetimibe), and an agent that inhibits microsomal triglyceride transfer protein (e.g. lomitapide).

[0081] As used herein, an " ANGPTL3 inhibitor" is any agent, which binds to or interacts with human ANGPTL3 and inhibits the normal biological function of ANGPTL3 *in vitro* or *in vivo*. Non-limiting examples of categories of ANGPTL3 inhibitors include small molecule ANGPTL3 antagonists, nucleic acid-based inhibitors of ANGPTL3 expression or activity (*e.g.*, siRNA or antisense), peptide-based molecules that specifically interact with ANGPTL3 (*e.g.*, peptibodies), receptor molecules that specifically interact with ANGPTL3, ANGPTL3-binding scaffold molecules (*e.g.*, DARPins, HEAT repeat proteins, ARM repeat proteins, tetratricopeptide repeat proteins, fibronectin-based scaffold constructs, and other scaffolds based on naturally occurring repeat proteins, etc., [*see, e.g.*, Boersma and Pluckthun, 2011,

Curr. Opin. Biotechnol. 22:849-857, and references cited therein]), and anti-ANGPTL3 aptamers or portions thereof. According to certain embodiments, ANGPTL3 inhibitors that can be used in the context of the present invention are anti-ANGPTL3 antibodies or antigen-binding fragments of antibodies that specifically bind human ANGPTL3.

[0082] The term "human angiopoietin-like protein-3" or "human ANGPTL3" or "hANGPTL3", as used herein, refers to ANGPTL3 having the amino acid sequence of SEQ ID NO: 9 (see also NCBI Accession NP_055310), or a biologically active fragment thereof.

[0083] The term "antibody", as used herein, is intended to refer to immunoglobulin molecules comprising four polypeptide chains, two heavy (H) chains and two light (L) chains inter-connected by disulfide bonds, as well as multimers thereof (*e.g.*, IgM). Each heavy chain comprises a heavy chain variable region (abbreviated herein as HCVR or V_H) and a heavy chain constant region. The heavy chain constant region comprises three domains, C_H1, C_H2 and C_H3. Each light chain comprises a light chain variable region (abbreviated herein as LCVR or V_L) and a light chain constant region. The light chain constant region comprises one domain (C_L1). The V_H and V_L regions can be further subdivided into regions of hypervariability, termed complementarity determining regions (CDRs), interspersed with regions that are more conserved, termed framework regions (FR). Each V_H and V_L is composed of three CDRs and four FRs, arranged from amino-terminus to carboxy-terminus in the following order: FR1, CDR1, FR2, CDR2, FR3, CDR3, FR4. In different embodiments of the invention, the FRs of the anti-ANGPTL3 antibody (or antigen-binding portion thereof) may be identical to the human germline sequences, or may be naturally or artificially modified. An amino acid consensus sequence may be defined based on a side-by-side analysis of two or more CDRs.

[0084] The term "antibody," as used herein, also includes antigen-binding fragments of full antibody molecules. The terms "antigen-binding portion" of an antibody, "antigen-binding fragment" of an antibody, and the like, as used herein, include any naturally occurring, enzymatically obtainable, synthetic, or genetically engineered polypeptide or glycoprotein that specifically binds an antigen to form a complex. Antigen-binding fragments of an antibody may be derived, *e.g.*, from full antibody molecules using any suitable standard techniques such as proteolytic digestion or recombinant genetic engineering techniques involving the manipulation and expression of DNA encoding antibody variable and optionally constant domains. Such DNA is known and/or is readily available from, *e.g.*, commercial sources, DNA libraries (including, *e.g.*, phage-antibody libraries), or can be synthesized. The DNA may be sequenced and manipulated chemically or by using molecular biology techniques, for example, to arrange one or more variable and/or constant domains into a suitable configuration, or to introduce codons, create cysteine residues, modify, add or delete amino acids, etc.

[0085] Non-limiting examples of antigen-binding fragments include: (i) Fab fragments; (ii) F(ab')2 fragments; (iii) Fd fragments; (iv) Fv fragments; (v) single-chain Fv (scFv)

molecules; (vi) dAb fragments; and (vii) minimal recognition units consisting of the amino acid residues that mimic the hypervariable region of an antibody (e.g., an isolated complementarity determining region (CDR) such as a CDR3 peptide), or a constrained FR3-CDR3-FR4 peptide. Other engineered molecules, such as domain-specific antibodies, single domain antibodies, domain-deleted antibodies, chimeric antibodies, CDR-grafted antibodies, diabodies, triabodies, tetrabodies, minibodies, nanobodies (e.g. monovalent nanobodies, bivalent nanobodies, etc.), small modular immunopharmaceuticals (SMIPs), and shark variable IgNAR domains, are also encompassed within the expression "antigen-binding fragment," as used herein.

[0086] An antigen-binding fragment of an antibody will typically comprise at least one variable domain. The variable domain may be of any size or amino acid composition and will generally comprise at least one CDR, which is adjacent to or in frame with one or more framework sequences. In antigen-binding fragments having a V_H domain associated with a V_L domain, the V_H and V_L domains may be situated relative to one another in any suitable arrangement. For example, the variable region may be dimeric and contain V_H-V_H, V_H-V_L or V_L-V_L dimers. Alternatively, the antigen-binding fragment of an antibody may contain a monomeric V_H or V_L domain.

[0087] In certain embodiments, an antigen-binding fragment of an antibody may contain at least one variable domain covalently linked to at least one constant domain. Non-limiting, exemplary configurations of variable and constant domains that may be found within an antigen-binding fragment of an antibody of the present invention include: (i) V_H-C_H1; (ii) V_H-C_H2; (iii) V_H-C_H3; (iv) V_H-C_H1-C_H2; (v) V_H-C_H1-C_H2-C_H3; (vi) V_H-C_H2-C_H3; (vii) V_H-C_L; (viii) V_L-C_H1; (ix) V_L-C_H2; (x) V_L-C_H3; (xi) V_L-C_H1-C_H2; (xii) V_L-C_H1-C_H2-C_H3; (xiii) V_L-C_H2-C_H3; and (xiv) V_L-C_L. In any configuration of variable and constant domains, including any of the exemplary configurations listed above, the variable and constant domains may be either directly linked to one another or may be linked by a full or partial hinge or linker region. A hinge region may consist of at least 2 (e.g., 5, 10, 15, 20, 40, 60 or more) amino acids, which result in a flexible or semi-flexible linkage between adjacent variable and/or constant domains in a single polypeptide molecule. Moreover, an antigen-binding fragment of an antibody of the present invention may comprise a homo-dimer or hetero-dimer (or other multimer) of any of the variable and constant domain configurations listed above in non-covalent association with one another and/or with one or more monomeric V_H or V_L domain (e.g., by disulfide bond(s)).

[0088] As with full antibody molecules, antigen-binding fragments may be monospecific or multispecific (e.g., bispecific). A multispecific antigen-binding fragment of an antibody will typically comprise at least two different variable domains, wherein each variable domain is capable of specifically binding to a separate antigen or to a different epitope on the same antigen. Any multispecific antibody format, including the exemplary bispecific antibody formats disclosed herein, may be adapted for use in the context of an antigen-binding fragment of an

antibody of the present invention using routine techniques available in the art.

[0089] The constant region of an antibody is important in the ability of an antibody to fix complement and mediate cell-dependent cytotoxicity. Thus, the isotype of an antibody may be selected on the basis of whether it is desirable for the antibody to mediate cytotoxicity.

[0090] The term "human antibody", as used herein, is intended to include antibodies having variable and constant regions derived from human germline immunoglobulin sequences. The human antibodies of the invention may nonetheless include amino acid residues not encoded by human germline immunoglobulin sequences (e.g., mutations introduced by random or site-specific mutagenesis *in vitro* or by somatic mutation *in vivo*), for example in the CDRs and in particular CDR3. However, the term "human antibody", as used herein, is not intended to include antibodies in which CDR sequences derived from the germline of another mammalian species, such as a mouse, have been grafted onto human framework sequences. The term includes antibodies recombinantly produced in a non-human mammal, or in cells of a non-human mammal. The term is not intended to include antibodies isolated from or generated in a human subject.

[0091] The term "recombinant human antibody", as used herein, is intended to include all human antibodies that are prepared, expressed, created or isolated by recombinant means, such as antibodies expressed using a recombinant expression vector transfected into a host cell (described further below), antibodies isolated from a recombinant, combinatorial human antibody library (described further below), antibodies isolated from an animal (e.g., a mouse) that is transgenic for human immunoglobulin genes (see e.g., Taylor et al. (1992) Nucl. Acids Res. 20:6287-6295) or antibodies prepared, expressed, created or isolated by any other means that involves splicing of human immunoglobulin gene sequences to other DNA sequences. Such recombinant human antibodies have variable and constant regions derived from human germline immunoglobulin sequences. In certain embodiments, however, such recombinant human antibodies are subjected to *in vitro* mutagenesis (or, when an animal transgenic for human Ig sequences is used, *in vivo* somatic mutagenesis) and thus the amino acid sequences of the V_H and V_L regions of the recombinant antibodies are sequences that, while derived from and related to human germline V_H and V_L sequences, may not naturally exist within the human antibody germline repertoire *in vivo*.

[0092] Human antibodies can exist in two forms that are associated with hinge heterogeneity. In one form, an immunoglobulin molecule comprises a stable four chain construct of approximately 150-160 kDa in which the dimers are held together by an interchain heavy chain disulfide bond. In a second form, the dimers are not linked via inter-chain disulfide bonds and a molecule of about 75-80 kDa is formed composed of a covalently coupled light and heavy chain (half-antibody). These forms have been extremely difficult to separate, even after affinity purification.

[0093] The frequency of appearance of the second form in various intact IgG isotypes is due to, but not limited to, structural differences associated with the hinge region isotype of the antibody. A single amino acid substitution in the hinge region of the human IgG4 hinge can significantly reduce the appearance of the second form (Angal et al. (1993) Molecular Immunology 30:105) to levels typically observed using a human IgG1 hinge. The instant invention encompasses antibodies having one or more mutations in the hinge, C_H2 or C_H3 region which may be desirable, for example, in production, to improve the yield of the desired antibody form.

[0094] An "isolated antibody," as used herein, means an antibody that has been identified and separated and/or recovered from at least one component of its natural environment. For example, an antibody that has been separated or removed from at least one component of an organism, or from a tissue or cell in which the antibody naturally exists or is naturally produced, is an "isolated antibody" for purposes of the present invention. An isolated antibody also includes an antibody *in situ* within a recombinant cell. Isolated antibodies are antibodies that have been subjected to at least one purification or isolation step. According to certain embodiments, an isolated antibody may be substantially free of other cellular material and/or chemicals.

[0095] The term "specifically binds," or the like, means that an antibody or antigen-binding fragment thereof forms a complex with an antigen that is relatively stable under physiologic conditions. Methods for determining whether an antibody specifically binds to an antigen are well known in the art and include, for example, equilibrium dialysis, surface plasmon resonance, and the like. For example, an antibody that "specifically binds" ANGPTL3, as used in the context of the present invention, includes antibodies that bind ANGPTL3, or a portion thereof with a K_D of less than about 1000 nM, less than about 500 nM, less than about 300 nM, less than about 200 nM, less than about 100 nM, less than about 90 nM, less than about 80 nM, less than about 70 nM, less than about 60 nM, less than about 50 nM, less than about 40 nM, less than about 30 nM, less than about 20 nM, less than about 10 nM, less than about 5 nM, less than about 4 nM, less than about 3 nM, less than about 2 nM, less than about 1 nM or less than about 0.5 nM, as measured in a surface plasmon resonance assay. An isolated antibody that specifically binds human ANGPTL3, however, has cross-reactivity to other antigens, such as ANGPTL3 molecules from other (non-human) species.

[0096] The anti-ANGPTL3 antibodies useful for the methods of the present invention may comprise one or more amino acid substitutions, insertions and/or deletions in the framework and/or CDR regions of the heavy and light chain variable domains as compared to the corresponding germline sequences from which the antibodies were derived. Such mutations can be readily ascertained by comparing the amino acid sequences disclosed herein to germline sequences available from, for example, public antibody sequence databases. The present invention includes methods involving the use of antibodies, and antigen-binding fragments thereof, which are derived from any of the amino acid sequences disclosed herein,

wherein one or more amino acids within one or more framework and/or CDR regions are mutated to the corresponding residue(s) of the germline sequence from which the antibody was derived, or to the corresponding residue(s) of another human germline sequence, or to a conservative amino acid substitution of the corresponding germline residue(s) (such sequence changes are referred to herein collectively as "germline mutations"). A person of ordinary skill in the art, starting with the heavy and light chain variable region sequences disclosed herein, can easily produce numerous antibodies and antigen-binding fragments which comprise one or more individual germline mutations or combinations thereof. In certain embodiments, all of the framework and/or CDR residues within the V_H and/or V_L domains are mutated back to the residues found in the original germline sequence from which the antibody was derived. In other embodiments, only certain residues are mutated back to the original germline sequence, *e.g.*, only the mutated residues found within the first 8 amino acids of FR1 or within the last 8 amino acids of FR4, or only the mutated residues found within CDR1, CDR2 or CDR3. In other embodiments, one or more of the framework and/or CDR residue(s) are mutated to the corresponding residue(s) of a different germline sequence (*i.e.*, a germline sequence that is different from the germline sequence from which the antibody was originally derived). Furthermore, the antibodies of the present invention may contain any combination of two or more germline mutations within the framework and/or CDR regions, *e.g.*, wherein certain individual residues are mutated to the corresponding residue of a particular germline sequence while certain other residues that differ from the original germline sequence are maintained or are mutated to the corresponding residue of a different germline sequence. Once obtained, antibodies and antigen-binding fragments that contain one or more germline mutations can be easily tested for one or more desired property such as, improved binding specificity, increased binding affinity, improved or enhanced antagonistic or agonistic biological properties (as the case may be), reduced immunogenicity, etc. The use of antibodies and antigen-binding fragments obtained in this general manner are encompassed within the present invention.

[0100] The present invention also includes methods involving the use of anti-ANGPTL3 antibodies comprising variants of any of the HCVR, LCVR, and/or CDR amino acid sequences disclosed herein having one or more conservative substitutions. For example, the present invention includes the use of anti-ANGPTL3 antibodies having HCVR, LCVR, and/or CDR amino acid sequences with, *e.g.*, 10 or fewer, 8 or fewer, 6 or fewer, 4 or fewer, etc. conservative amino acid substitutions relative to any of the HCVR, LCVR, and/or CDR amino acid sequences disclosed herein.

[0101] The term "surface plasmon resonance", as used herein, refers to an optical phenomenon that allows for the analysis of real-time interactions by detection of alterations in protein concentrations within a biosensor matrix, for example using the BIAcore™ system (Biacore Life Sciences division of GE Healthcare, Piscataway, NJ).

[0102] The term "K_D ", as used herein, is intended to refer to the equilibrium dissociation

constant of a particular antibody-antigen interaction.

[0103] The term "epitope" refers to an antigenic determinant that interacts with a specific antigen binding site in the variable region of an antibody molecule known as a paratope. A single antigen may have more than one epitope. Thus, different antibodies may bind to different areas on an antigen and may have different biological effects. Epitopes may be either conformational or linear. A conformational epitope is produced by spatially juxtaposed amino acids from different segments of the linear polypeptide chain. A linear epitope is one produced by adjacent amino acid residues in a polypeptide chain. In certain circumstance, an epitope may include moieties of saccharides, phosphoryl groups, or sulfonyl groups on the antigen.

[0104] According to certain embodiments, the anti-ANGPTL3 antibodies used in the methods of the present invention are antibodies with pH-dependent binding characteristics. As used herein, the expression "pH-dependent binding" means that the antibody or antigen-binding fragment thereof exhibits "reduced binding to ANGPTL3 at acidic pH as compared to neutral pH" (for purposes of the present disclosure, both expressions may be used interchangeably). For the example, antibodies "with pH-dependent binding characteristics" includes antibodies and antigen-binding fragments thereof that bind to ANGPTL3 with higher affinity at neutral pH than at acidic pH. In certain embodiments, the antibodies and antigen-binding fragments of the present invention bind ANGPTL3 with at least 3, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, or more times higher affinity at neutral pH than at acidic pH.

[0105] According to this aspect of the invention, the anti-ANGPTL3 antibodies with pH-dependent binding characteristics may possess one or more amino acid variations relative to the parental anti-ANGPTL3 antibody. For example, an anti-ANGPTL3 antibody with pH-dependent binding characteristics may contain one or more histidine substitutions or insertions, e.g., in one or more CDRs of a parental anti-ANGPTL3 antibody. Thus, according to certain embodiments of the present invention, methods are provided comprising administering an anti-ANGPTL3 antibody which comprises CDR amino acid sequences (e.g., heavy and light chain CDRs) which are identical to the CDR amino acid sequences of a parental ANGPTL3 antibody except for the substitution of one or more amino acids of one or more CDRs of the parental antibody with a histidine residue. The anti-ANGPTL3 antibodies with pH-dependent binding may possess, e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, or more histidine substitutions, either within a single CDR of a parental antibody or distributed throughout multiple (e.g., 2, 3, 4, 5, or 6) CDRs of a parental anti-ANGPTL3 antibody. For example, the present invention includes the use of anti-ANGPTL3 antibodies with pH-dependent binding comprising one or more histidine substitutions in HCDR1, one or more histidine substitutions in HCDR2, one or more histidine substitutions in HCDR3, one or more histidine substitutions in LCDR1, one or more histidine substitutions in LCDR2, and/or one or more histidine substitutions in LCDR3, of a parental anti-ANGPTL3 antibody.

[0106] As used herein, the expression "acidic pH" means a pH of 6.0 or less (*e.g.*, less than about 6.0, less than about 5.5, less than about 5.0, etc.). The expression "acidic pH" includes pH values of about 6.0, 5.95, 5.90, 5.85, 5.8, 5.75, 5.7, 5.65, 5.6, 5.55, 5.5, 5.45, 5.4, 5.35, 5.3, 5.25, 5.2, 5.15, 5.1, 5.05, 5.0, or less. As used herein, the expression "neutral pH" means a pH of about 7.0 to about 7.4. The expression "neutral pH" includes pH values of about 7.0, 7.05, 7.1, 7.15, 7.2, 7.25, 7.3, 7.35, and 7.4.

[0107] A non-limiting example of an anti-ANGPTL3 antibody that can be used in the context of the present invention includes evinacumab.

Preparation of Human Antibodies

[0108] Anti-ANGPTL3 antibodies can be made according to any method of antibody production/isolation known in the art. For example, antibodies for use in the methods of the present invention may be made by hybridoma technologies, by phage display, by yeast display, etc. Antibodies for use in the methods of the present invention may be, *e.g.*, chimeric antibodies, humanized antibodies, or fully human antibodies.

[0109] Methods for generating human antibodies in transgenic mice are known in the art. Any such known methods can be used in the context of the present invention to make human antibodies that specifically bind ANGPTL3.

[0110] For example, using VELOCIMMUNE™ technology (see, for example, US 6,596,541, Regeneron Pharmaceuticals) or any other known method for generating monoclonal antibodies, high affinity chimeric antibodies to ANGPTL3 are initially isolated having a human variable region and a mouse constant region. The VELOCIMMUNE® technology involves generation of a transgenic mouse having a genome comprising human heavy and light chain variable regions operably linked to endogenous mouse constant region loci such that the mouse produces an antibody comprising a human variable region and a mouse constant region in response to antigenic stimulation. The DNA encoding the variable regions of the heavy and light chains of the antibody are isolated and operably linked to DNA encoding the human heavy and light chain constant regions. The DNA is then expressed in a cell capable of expressing the fully human antibody.

[0111] Generally, a VELOCIMMUNE® mouse is challenged with the antigen of interest, and lymphatic cells (such as B-cells) are recovered from the mice that express antibodies. The lymphatic cells may be fused with a myeloma cell line to prepare immortal hybridoma cell lines, and such hybridoma cell lines are screened and selected to identify hybridoma cell lines that produce antibodies specific to the antigen of interest. DNA encoding the variable regions of the heavy chain and light chain may be isolated and linked to desirable isotypic constant regions of the heavy chain and light chain. Such an antibody protein may be produced in a cell, such as a CHO cell. Alternatively, DNA encoding the antigen-specific chimeric antibodies or the variable domains of the light and heavy chains may be isolated directly from antigen-specific

lymphocytes.

[0112] Initially, high affinity chimeric antibodies are isolated having a human variable region and a mouse constant region. The antibodies are characterized and selected for desirable characteristics, including affinity, selectivity, epitope, etc., using standard procedures known to those skilled in the art. The mouse constant regions are replaced with a desired human constant region to generate the fully human antibody of the invention, for example wild-type or modified IgG1 or IgG4. While the constant region selected may vary according to specific use, high affinity antigen-binding and target specificity characteristics reside in the variable region.

[0113] In general, the antibodies that can be used in the methods of the present invention possess high affinities, as described above, when measured by binding to antigen either immobilized on solid phase or in solution phase. The mouse constant regions are replaced with desired human constant regions to generate the fully human antibodies of the invention. While the constant region selected may vary according to specific use, high affinity antigen-binding and target specificity characteristics reside in the variable region.

[0114] Specific examples of human antibodies or antigen-binding fragments of antibodies that specifically bind ANGPTL3, which can be used in the context of the methods of the present invention include antibodies or antigen-binding proteins comprising the six CDRs (HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3) from the heavy and light chain variable region (HCVR/LCVR) amino acid sequence pair comprising SEQ ID NOs: 1/5.

[0115] In certain embodiments of the present invention, the anti-ANGPTL3 antibody, or antigen-binding fragment thereof, that can be used in the methods of the present invention comprises heavy and light chain complementarity determining regions (HCDR1-HCDR2-HCDR3/LCDR1-LCDR2-LCDR3) comprising the amino acid sequences of SEQ ID NOs:2, 3, 4, 6, 7 and 8.

[0116] In certain embodiments of the present invention, the anti-ANGPTL3 antibody, or antigen-binding fragment thereof, that can be used in the methods of the present invention comprises an HCVR having the amino acid sequence of SEQ ID NO:1 and an LCVR having the amino acid sequence of SEQ ID NO:5.

Pharmaceutical Compositions and Methods of Administration

[0117] The present invention includes methods, which comprise administering an ANGPTL3 inhibitor to a patient in combination with a statin, an inhibitor of cholesterol absorption and an inhibitor of microsomal triglyceride transfer protein, wherein the ANGPTL3 inhibitor and the additional agents are contained within the same, or in different pharmaceutical compositions. The pharmaceutical compositions of the invention are formulated with suitable carriers, excipients, and other agents that provide suitable transfer, delivery, tolerance, and the like. A multitude of appropriate formulations can be found in the formulary known to all pharmaceutical chemists: Remington's Pharmaceutical Sciences, Mack Publishing Company, Easton, PA.

These formulations include, for example, powders, pastes, ointments, jellies, waxes, oils, lipids, lipid (cationic or anionic) containing vesicles (such as LIPOFECTIN™), DNA conjugates, anhydrous absorption pastes, oil-in-water and water-in-oil emulsions, emulsions carbowax (polyethylene glycols of various molecular weights), semi-solid gels, and semi-solid mixtures containing carbowax. See also Powell et al. "Compendium of excipients for parenteral formulations" PDA (1998) J Pharm Sci Technol 52:238-311.

[0118] Exemplary pharmaceutical formulations comprising anti-ANGPTL3 antibodies that can be used in the context of the present invention include any of the formulations as set forth in US 8,795,669 (describing, *inter alia*, exemplary formulations comprising alirocumab), or in WO2013/166448, or WO2012/168491.

[0119] Various delivery systems are known and can be used to administer the pharmaceutical composition of the invention, *e.g.*, encapsulation in liposomes, microparticles, microcapsules, recombinant cells capable of expressing the mutant viruses, receptor mediated endocytosis (see, e.g., Wu et al., 1987, J. Biol. Chem. 262:4429-4432). Methods of administration include, but are not limited to, intradermal, intramuscular, intraperitoneal, intravenous, subcutaneous, intranasal, epidural, and oral routes. The composition may be administered by any convenient route, for example by infusion or bolus injection, by absorption through epithelial or mucocutaneous linings (e.g., oral mucosa, rectal and intestinal mucosa, etc.) and may be administered together with other biologically active agents.

[0120] A pharmaceutical composition of the present invention can be delivered subcutaneously or intravenously with a standard needle and syringe. In addition, with respect to subcutaneous delivery, a pen delivery device readily has applications in delivering a pharmaceutical composition of the present invention. Such a pen delivery device can be reusable or disposable. A reusable pen delivery device generally utilizes a replaceable cartridge that contains a pharmaceutical composition. Once all of the pharmaceutical composition within the cartridge has been administered and the cartridge is empty, the empty cartridge can readily be discarded and replaced with a new cartridge that contains the pharmaceutical composition. The pen delivery device can then be reused. In a disposable pen delivery device, there is no replaceable cartridge. Rather, the disposable pen delivery device comes prefilled with the pharmaceutical composition held in a reservoir within the device. Once the reservoir is emptied of the pharmaceutical composition, the entire device is discarded.

[0121] Numerous reusable pen and autoinjector delivery devices have applications in the subcutaneous delivery of a pharmaceutical composition of the present invention. Examples include, but are not limited to AUTOPEN™ (Owen Mumford, Inc., Woodstock, UK), DISETRONIC™ pen (Disetronic Medical Systems, Bergdorf, Switzerland), HUMALOG MIX 75/25™ pen, HUMALOG™ pen, HUMALIN 70/30™ pen (Eli Lilly and Co., Indianapolis, IN), NOVOPEN™ I, II and III (Novo Nordisk, Copenhagen, Denmark), NOVOPEN JUNIOR™ (Novo

Nordisk, Copenhagen, Denmark), BD™ pen (Becton Dickinson, Franklin Lakes, NJ),
OPTIPEN™, OPTIPEN PRO™, OPTIPEN STARLET™, and OPTICLIK™ (Sanofi-Aventis,
Frankfurt, Germany), to name only a few. Examples of disposable pen delivery devices having
applications in subcutaneous delivery of a pharmaceutical composition of the present invention
include, but are not limited to the SOLOSTAR™ pen (Sanofi-Aventis), the FLEXPEN™ (Novo
Nordisk), and the KWIKPEN™ (Eli Lilly), the SURECLICK™ Autoinjector (Amgen, Thousand
Oaks, CA), the PENLET™ (Haselmeier, Stuttgart, Germany), the EPIPEN (Dey, L.P.), and the
HUMIRA™ Pen (Abbott Labs, Abbott Park IL), to name only a few.

[0122] In certain situations, the pharmaceutical composition can be delivered in a controlled
release system. In one embodiment, a pump may be used (see Langer, supra; Sefton, 1987,
CRC Crit. Ref. Biomed. Eng. 14:201). In another embodiment, polymeric materials can be
used; see, Medical Applications of Controlled Release, Langer and Wise (eds.), 1974, CRC
Pres., Boca Raton, Florida. In yet another embodiment, a controlled release system can be
placed in proximity of the composition's target, thus requiring only a fraction of the systemic
dose (see, *e.g.*, Goodson, 1984, in Medical Applications of Controlled Release, supra, vol. 2, pp.
115-138). Other controlled release systems are discussed in the review by Langer, 1990,
Science 249:1527-1533.

[0123] The injectable preparations may include dosage forms for intravenous, subcutaneous,
intracutaneous and intramuscular injections, drip infusions, etc. These injectable preparations
may be prepared by known methods. For example, the injectable preparations may be
prepared, *e.g.*, by dissolving, suspending or emulsifying the antibody or its salt described above
in a sterile aqueous medium or an oily medium conventionally used for injections. As the
aqueous medium for injections, there are, for example, physiological saline, an isotonic solution
containing glucose and other auxiliary agents, etc., which may be used in combination with an
appropriate solubilizing agent such as an alcohol (e.g., ethanol), a polyalcohol (e.g., propylene
glycol, polyethylene glycol), a nonionic surfactant [e.g., polysorbate 80, HCO-50
(polyoxyethylene (50 mol) adduct of hydrogenated castor oil)], etc. As the oily medium, there
are employed, e.g., sesame oil, soybean oil, etc., which may be used in combination with a
solubilizing agent such as benzyl benzoate, benzyl alcohol, etc. The injection thus prepared is
preferably filled in an appropriate ampoule.

[0124] Advantageously, the pharmaceutical compositions for oral or parenteral use described
above are prepared into dosage forms in a unit dose suited to fit a dose of the active
ingredients. Such dosage forms in a unit dose include, for example, tablets, pills, capsules,
injections (ampoules), suppositories, etc.

Dosage

[0125] The amount of an ANGPTL3 inhibitor (*e.g.*, anti-ANGPTL3 antibody) administered to a
subject according to the methods of the present invention is, generally, a therapeutically

effective amount. As used herein, the phrase "therapeutically effective amount of an ANGPTL3 inhibitor" means a dose of an ANGPTL3 inhibitor, when administered in combination with a statin, ezetimibe and lomitapide, results in a detectable reduction (at least about 5%, 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, or more from baseline) in one or more parameters selected from the group consisting of total cholesterol, LDL-C, ApoB, ApoA-1, Apo CIII, non-HDL-C, VLDL-C, triglycerides, and Lp(a), or an amount that reduces or eliminates a patient's need for other therapeutic agents, or interventions, such as, for example, lipoprotein apheresis.

[0126] The amount of ANGPTL3 inhibitor (e.g., anti-ANGPTL3 antibody) administered to a subject according to the methods of the present invention is, generally, a therapeutically effective amount. As used herein, the phrase "therapeutically effective amount of an ANGPTL3 inhibitor" means a dose of ANGPTL3 inhibitor, when combined with the therapeutic agents described herein, results in a detectable reduction (at least about 5%, 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, or more from baseline) in one or more parameters selected from the group consisting of total cholesterol, LDL-C, ApoB, ApoA-1, Apo CIII, non-HDL-C, VLDL-C, triglycerides, and Lp(a).

[0127] In the case of an anti-ANGPTL3 antibody, a therapeutically effective amount can be from about 0.05 mg to about 600 mg, e.g., about 0.05 mg, about 0.1 mg, about 1.0 mg, about 1.5 mg, about 2.0 mg, about 10 mg, about 20 mg, about 30 mg, about 40 mg, about 50 mg, about 60 mg, about 70 mg, about 80 mg, about 90 mg, about 100 mg, about 110 mg, about 120 mg, about 130 mg, about 140 mg, about 160 mg, about 170 mg, about 180 mg, about 190 mg, about 200 mg, about 210 mg, about 220 mg, about 230 mg, about 240 mg, about 250 mg, about 260 mg, about 270 mg, about 280 mg, about 290 mg, about 300 mg, about 310 mg, about 320 mg, about 330 mg, about 340 mg, about 350 mg, about 360 mg, about 370 mg, about 380 mg, about 390 mg, about 400 mg, about 410 mg, about 420 mg, about 430 mg, about 440 mg, about 450 mg, about 460 mg, about 470 mg, about 480 mg, about 490 mg, about 500 mg, about 510 mg, about 520 mg, about 530 mg, about 540 mg, about 550 mg, about 560 mg, about 570 mg, about 580 mg, about 590 mg, or about 600 mg, of the anti-ANGPTL3 antibody. Other dosing amounts of ANGPTL3 inhibitors will be apparent to persons of ordinary skill in the art and are contemplated within the scope of the present invention.

[0128] The amount of anti-ANGPTL3 antibody contained within the individual doses may be expressed in terms of milligrams of antibody per kilogram of patient body weight (i.e., mg/kg). For example, the anti-ANGPTL3 antibody may be administered to a patient at a dose of about 0.0001 to about 20 mg/kg of patient body weight.

Combination Therapies

[0129] The methods of the present invention may also comprise administering an ANGPTL3 inhibitor in combination with a statin, ezetimibe and lomitapide to a patient who is non-

responsive to, inadequately controlled by, or intolerant to other standard lipid lowering therapies. In certain embodiments, the need for further administration of the standard lipid lowering therapy may be eliminated altogether. In certain embodiments, the combined use of the ANGPTL3 inhibitor with the other agents described herein may be used in combination with ("on top of") the patient's previously prescribed lipid lowering therapy. For example, in the context of lowering at least one lipid/lipoprotein parameter in a patient suffering from hyperlipidemia (e.g. hypercholesterolemia), wherein the patient is non-responsive to, inadequately controlled by, or intolerant to a standard lipid lowering therapy, a combination of an ANGPTL3 inhibitor with ezetimibe and lomitapide may be administered to a patient in combination with a stable daily therapeutic statin regimen. Exemplary daily therapeutic statin regimens that may be used in the context of the present invention, include, e.g., atorvastatin (10, 20, 40 or 80 mg daily), (atorvastatin/ezetimibe 10/10 or 40/10 mg daily), rosuvastatin (5, 10 or 20 mg daily), cerivastatin (0.4 or 0.8 mg daily), pitavastatin (1, 2 or 4 mg daily), fluvastatin (20, 40 or 80 mg daily), simvastatin (5, 10, 20, 40 or 80 mg daily), simvastatin/ezetimibe (10/10, 20/10, 40/10 or 80/10 mg daily), lovastatin (10, 20, 40 or 80 mg daily), pravastatin (10, 20, 40 or 80 mg daily), and combinations thereof. Other lipid modifying therapies that an ANGPTL3 inhibitor may be administered in combination with in the context of the present invention include, e.g., (1) an agent which increase lipoprotein catabolism (such as niacin); and/or (2) activators of the LXR transcription factor that plays a role in cholesterol elimination such as 22-hydroxycholesterol.

[0130] A non-limiting example of an ANGPTL3 antibody to be used in the context of the present invention includes evinacumab.

Administration Regimens

[0131] According to certain embodiments of the present invention, multiple doses of an ANGPTL3 inhibitor (i.e., a pharmaceutical composition comprising an ANGPTL3 inhibitor) may be administered to a subject over a defined time course (e.g., on top of a daily therapeutic statin regimen or other background lipid modifying therapy), in addition to administration of ezetimibe and lomitapide. The methods according to this aspect of the invention comprise sequentially administering to a subject multiple doses of an ANGPTL3 inhibitor. As used herein, "sequentially administering" means that each dose of ANGPTL3 inhibitor is administered to the subject at a different point in time, e.g., on different days separated by a predetermined interval (e.g., hours, days, weeks or months). The present invention includes methods which comprise sequentially administering to the patient a single initial dose of an ANGPTL3 inhibitor, followed by one or more secondary doses of the ANGPTL3 inhibitor, and optionally followed by one or more tertiary doses of the ANGPTL3 inhibitor.

[0132] The terms "initial dose," "secondary doses," and "tertiary doses," refer to the temporal sequence of administration of the individual doses of a pharmaceutical composition comprising

a ANGPTL3 inhibitor. Thus, the "initial dose" is the dose which is administered at the beginning of the treatment regimen (also referred to as the "baseline dose"); the "secondary doses" are the doses which are administered after the initial dose; and the "tertiary doses" are the doses which are administered after the secondary doses. The initial, secondary, and tertiary doses may all contain the same amount of the ANGPTL3 inhibitor, but generally may differ from one another in terms of frequency of administration. In certain embodiments, however, the amount of the ANGPTL3 inhibitor contained in the initial, secondary and/or tertiary doses varies from one another (*e.g.*, adjusted up or down as appropriate) during the course of treatment. In certain embodiments, two or more (*e.g.*, 2, 3, 4, or 5) doses are administered at the beginning of the treatment regimen as "loading doses" followed by subsequent doses that are administered on a less frequent basis (*e.g.*, "maintenance doses").

[0133] According to exemplary embodiments of the present invention, each secondary and/or tertiary dose is administered 1 to 26 (*e.g.*, 1, 1½, 2, 2½, 3, 3½, 4, 4½, 5, 5½, 6, 6½, 7, 7½, 8, 8½, 9, 9½, 10, 10½, 11, 11½, 12, 12½, 13, 13½, 14, 14½, 15, 15½, 16, 16½, 17, 17½, 18, 18½, 19, 19½, 20, 20½, 21, 21½, 22, 22½, 23, 23½, 24, 24½, 25, 25½, 26, 26½, or more) weeks after the immediately preceding dose. The phrase "the immediately preceding dose," as used herein, means, in a sequence of multiple administrations, the dose of antigen-binding molecule, which is administered to a patient prior to the administration of the very next dose in the sequence with no intervening doses.

[0134] The methods according to this aspect of the invention may comprise administering to a patient any number of secondary and/or tertiary doses of an ANGPTL3 inhibitor. For example, in certain embodiments, only a single secondary dose is administered to the patient. In other embodiments, two or more (*e.g.*, 2, 3, 4, 5, 6, 7, 8, or more) secondary doses are administered to the patient. Likewise, in certain embodiments, only a single tertiary dose is administered to the patient. In other embodiments, two or more (*e.g.*, 2, 3, 4, 5, 6, 7, 8, or more) tertiary doses are administered to the patient.

[0135] In embodiments involving multiple secondary doses, each secondary dose may be administered at the same frequency as the other secondary doses. For example, each secondary dose may be administered to the patient 1 to 2, 4, 6, 8 or more weeks after the immediately preceding dose. Similarly, in embodiments involving multiple tertiary doses, each tertiary dose may be administered at the same frequency as the other tertiary doses. For example, each tertiary dose may be administered to the patient 1 to 2, 4, 6, 8 or more weeks after the immediately preceding dose. Alternatively, the frequency at which the secondary and/or tertiary doses are administered to a patient can vary over the course of the treatment regimen. The frequency of administration may also be adjusted during the course of treatment by a physician depending on the needs of the individual patient following clinical examination. Likewise, the doses of the concomitant therapies, e.g. the statin, ezetimibe and lomitapide may be adjusted during the course of treatment by a physician according to the normalization of lipid

levels observed during the course of treatment.

EXAMPLES

[0136] The following examples are put forth so as to provide those of ordinary skill in the art with a complete disclosure and description of how to make and use the methods and compositions of the invention, and are not intended to limit the scope of what the inventors regard as their invention. Efforts have been made to ensure accuracy with respect to numbers used (e.g., amounts, temperature, etc.) but some experimental errors and deviations should be accounted for. Unless indicated otherwise, parts are parts by weight, molecular weight is average molecular weight, temperature is in degrees Centigrade, and pressure is at or near atmospheric.

Example 1. Generation of Human Antibodies to Human ANGPTL3

[0137] The exemplary ANGPTL3 antibody used in the following Example is the human anti-ANGPTL3 antibody known as "evinacumab." Evinacumab has the following amino acid sequence characteristics: a heavy chain variable region (HCVR) comprising SEQ ID NO:1 and a light chain variable domain (LCVR) comprising SEQ ID NO:5; a heavy chain complementarity determining region 1 (HCDR1) comprising SEQ ID NO:2, a HCDR2 comprising SEQ ID NO:3, a HCDR3 comprising SEQ ID NO:4, a light chain complementarity determining region 1 (LCDR1) comprising SEQ ID NO:6, a LCDR2 comprising SEQ ID NO:7 and a LCDR3 comprising SEQ ID NO:8.

Example 2: Safety and Efficacy of Evinacumab, a Monoclonal Antibody to ANGPTL3 in Patients with Homozygous Familial Hypercholesterolemia Receiving Concomitant Lipid-lowering Therapies

[0138] Homozygous familial hypercholesterolemia (HoFH) involves profound genetic deficiencies in the low-density lipoprotein (LDL) receptor pathway, leading to catastrophically elevated LDL-cholesterol (LDL-C) and severe premature atherosclerosis; responses to statins and PCSK9 antibodies are limited. Preclinical studies and human genetic analyses suggest that inhibition of angiopoietin-like protein (ANGPTL3) lowers LDL-C and provides cardiovascular benefit, independently of the LDL receptor. Evinacumab, a human ANGPTL3 antibody, was administered to nine HoFH adults (three null homozygotes) already on maximally-tolerated conventional therapies. LDL-C decreased 49% (range –25% to –90%) at week 4 (primary endpoint). Overall mean peak reduction in LDL-C was –58±18% (-90% to -33%) between weeks 4 and 12, showing that ANGPTL3 inhibition by evinacumab substantially reduces LDL-C in HoFH patients.

[0139] Low-density lipoproteins (LDL) play a major role in the initiation and progression of atherosclerosis and risk of cardiovascular disease. Familial hypercholesterolemia (FH) is typically a disorder occurring through mutations in genes encoding proteins that regulate clearance of LDL. These include the genes for the low-density lipoprotein receptor (*LDLR*), apolipoprotein B (*APOB*), proprotein convertase subtilisin/kexin type 9 (*PCSK9*), and low-density lipoprotein receptor adaptor protein 1 (*LDLRAP1*) (Cuchel, *et al.* 2014 *Eur Heart J* 35:2146-57). Patients with heterozygous FH usually have untreated plasma LDL-cholesterol (LDL-C) levels ranging from 350 to 550 mg per deciliter and generally respond to lipid-lowering therapies, including moderate to high doses of high-potency statins, ezetimibe, and PCSK9 antibodies (Goldberg, *et al.* 2011 *J Clin Lipidol* 2011;5:S1-8; Kastelein, *et al.* 2014 *Cardiovasc Drugs Ther* 28:281-9). Homozygous FH (HoFH) is a rare disease, which affects 1 in 160,000 to 300,000 people. Patients with HoFH carry two FH-causing mutations (homozygous or compound heterozygous), have much higher untreated LDL-C levels, generally ranging from 500 to 1000 mg per deciliter (Kolansky, *et al.* 2008 *The American journal of cardiology* 102:1438-43), and are significantly less responsive or unresponsive to standard lipid-lowering therapies. Most individuals with HoFH develop severe xanthomatosis, coronary heart disease, and peripheral atherosclerosis at an early age, and can die before the age of 30 if left untreated (Nordestgaard, *et al.* 2013 *Eur Heart J* 34:3478-90a).

[0140] Genetic and phenotypic heterogeneity in HoFH can translate into broad variability in cardiovascular disease manifestation and response to lipid-lowering therapies. Some FH-causing mutations result in defective LDL receptors with residual activity, whereas others have no activity and therefore do not respond to conventional lipid-lowering medications, such as statins and PCSK9 antibodies, which mainly target the process of LDL-receptor expression (Santos PC, Pereira AC. 2015 *Pharmacogenomics* 16:1743-50; Rader DJ, Kastelein JJ. 2014 *Circulation* 129:1022-32). Drugs with mechanisms of action unrelated to the LDL receptor, such as lomitapide and mipomersen, have been approved recently for treating HoFH, but their use can be hampered by tolerability and safety issues.

[0141] Angiopoietin-like protein 3 (ANGPTL3) is a secreted protein expressed in the liver. It acts to increase plasma levels of triglycerides, LDL-C, and high-density lipoprotein cholesterol (HDL-C) by inhibiting the activity of lipoprotein lipase and endothelial lipase or by modulating the clearance of triglyceride-rich lipoproteins upstream of LDL production (Wang, *et al.* 2015 *J Lipid Res* 56:1296-307; Musunuru, *et al.* 2010 *N Engl J Med* 363:2220-7). Preclinical studies show that knockout of ANGPTL3, or blockade by an antibody, can lower triglycerides and LDL-C independently of the LDL-R, and have benefit in models of atherosclerosis (Ando, *et al.* 2003 *J Lipid Res* 44:1216-23; Dewey, *et al.* 2017 *New Engl J Med*; in press). Consistent with this, large-scale genetic studies in man show that loss-of-function mutations in *ANGPTL3* lead to reduced plasma levels of triglycerides, LDL-C, and HDL-C (Robciuc, *et al.* 2013

Arteriosclerosis, thrombosis, and vascular biology 33:1706-13; Pisciotta, *et al.* 2012 *Circ Cardiovasc Genet* 5:42-50; Minicocci, *et al.* 2013 *J Lipid Res* 54:3481-90; Wang, *et al.* 2015 *Proc Natl Acad Sci U S A* 112:11630-5; Noto, *et al.* 2012 *Arteriosclerosis, thrombosis, and vascular biology* 32:805-9; Martin-Campos, *et al.* 2012 *Clinica chimica acta; international journal of clinical chemistry* 413:552-5), and even more importantly, that these lipid changes associated with *ANGPTL3* mutations are also associated with protection from cardiovascular disease (Stitziel, *et al.* 2017 *Journal of the American College of Cardiology*). Altogether, the preclinical studies as well as the human genetic analyses suggest that ANGPTL3-inhibiting therapies could reduce LDL-C and provide benefit in patients with FH, including those suffering from profound homozygous disease, Evinacumab is a fully human monoclonal antibody that specifically blocks ANGPTL3 (Gusarova, *et al.* 2015 *J Lipid Res* 56:1308-17). In normal healthy volunteers, evinacumab was well tolerated and reduced the three major lipid fractions. A phase 2 study was conducted to determine whether evinacumab reduced LDL-C levels in nine patients with genetically and phenotypically confirmed HoFH, including patients homozygous for null mutations completely lacking LDLR activity.

METHODS

[0142] Patients: The nine patients (5 men, 4 women) were selected based on their genotypes and phenotypes. All presented a history of LDL-C >500 mg per deciliter or >400 mg per deciliter after portacaval shunt, premature atherosclerosis (8 of 9 with prior history of cardiovascular events) and severe xanthomatosis, and were homozygotes or compound heterozygotes for known FH-causing *LDLR* mutations (Hobbs, *et al.* 1992 *Hum Mutat* 1:445-66). Three patients were null homozygotes. All patients were on maximally tolerated lipid-lowering therapy.

[0143] Study Treatment: The patients were required to maintain their usual background lipid-lowering therapy and diet and exercise regimens throughout the study. All patients received a single open-label dose of evinacumab 250 mg subcutaneously in the abdominal area during the baseline visit and a single 15 mg per kilogram intravenous dose of evinacumab 2 weeks later. The sterile, lyophilized evinacumab drug product was supplied in a 5-ml single-use glass vial for reconstitution to a concentration of 100 mg per milliliter for subcutaneous doses and 50 mg per milliliter for intravenous doses. Patients were followed for a period of up to 24 weeks after the intravenous dose to allow for washout of evinacumab, and were offered enrollment in an extension study.

[0144] Pharmacodynamic Assessment: Fasting blood samples were collected before administration of study drug, at baseline and at regular intervals during the open-label treatment period and safety follow-up period, for measurement of LDL-C, non–HDL-C, total cholesterol, HDL-C, apolipoprotein B, lipoprotein(a), triglycerides, apolipoprotein A-1, and other parameters. The primary endpoint was the mean ± standard deviation (SD) percent change in LDL-C from baseline to week 4.

RESULTS

[0145] Even though most patients were on maximally tolerated therapies, mean±SD baseline LDL-C was 376.0±240.9 milligrams per deciliter (mg/dL); one patient who had failed statin therapy, and was removed from weekly apheresis, had a baseline LDL-C of 756 mg/dL. All nine patients reported the occurrence of at least one adverse event, but none led to treatment discontinuation. One event (coronary artery disease due to underlying disease) was serious but was not considered as related to the study medication. Six events were considered as related to the study medication, two of which were injection site reactions of mild severity, one was myalgia of moderate severity, and one was epistaxis of severe severity.

[0146] Drug Response: The mean±SD percent change in LDL-C from baseline to week 4 (pre-specified primary endpoint) following evinacumab administration was −49±23% (range: −90% to −25%), with an absolute change from baseline of −157±90 (range: −323 to −71) mg per deciliter (Table 1, below). The mean±SD achieved LDL-C value at week 4 was 219±191 mg per deciliter. Over the same period, percent change in apolipoprotein B decreased by 46±18% (Table 2, below), non-HDL-C by 49±22% (Table 3, below), triglycerides by 47% (median, interquartile range -57% to -38%), and HDL-C by 36±16%. Overall mean±SD peak reduction in LDL-C occurring between weeks 4 and 12 was −58±18% (range -90% to -33%), with an absolute peak reduction in LDL-C of 202 mg/dL. At week 4 (2 weeks after the intravenous dose), one patient achieved a reduction in LDL-C greater than 80%. In the 3 homozygous null patients, the mean±SD peak reduction in LDL-C through week 12 was -48±13% (range -60% to -33%).

Table 1: Effect of AngPTL3 Inhibition on Plasma LDL-C Concentrations

LDL-C	
Visit	Mean of Percent Change from Baseline ± SE (%)
BL (wk 0)	0
Day 4	-12.8 ± 3.7
Week 1	-24 ± 7.0
Week 2	-30.2 ± 8.1
Week 3	-41.4 ± 8.3
Week 4	-49.2 ± 7.7
Week 5	-46.8 ± 5.1
Week 6	-52.1 ± 4.9
Week 8	-51.6 ± 6.0
Week 10	-45.6 ± 4.6
Week 12	-36.6 ± 6.4

Table 2: Effect of AngPTL3 Inhibition on Apolipoprotein B Concentrations

Apolipoprotein B	
Visit	Mean of Percent Change from Baseline ± SE (%)
BL (wk 0)	0
Week 2	-24.2 ± 7
Week 3	-38.6 ± 7.3
Week 4	-45.9 ± 6.1
Week 5	-42.3 ± 4.7
Week 6	-43.1 ± 4.9
Week 8	-42.7 ± 4.8
Week 12	-29.5 ± 7.2

Table 3: Effect of AngPTL3 Inhibition on Non-HDL-C Concentrations

Non-HDL-C	
Visit	Mean of Percent Change from Baseline ± SE (%)
BL (wk 0)	0
Day 4	-13.6 ± 3.6
Week 1	-24.1 ± 6.7
Week 2	-29.6 ± 7.8
Week 3	-41.6 ± 8
Week 4	-48.9 ± 7.4
Week 5	-46.6 ± 5.0
Week 6	-51.5 ± 4.8
Week 8	-50.6 ± 5.7
Week 10	-44.8 ± 4.4
Week 12	-36.4 ± 6.2

[0147] Administration of the fully human monoclonal ANGPTL3-blocking antibody evinacumab in nine adults with HoFH, including three null homozygotes, resulted in meaningful reductions in LDL-C. Importantly, these reductions were on top of baseline levels already achieved on stable, maximally-tolerated lipid-lowering therapy with or without lomitapide, PCSK9 monoclonal antibodies or portacaval shunt. These results provide proof-of-concept of a substantial additional reduction in LDL-C by evinacumab, on top of standard of care, in the treatment of HoFH, with the potential for LDL-C normalization in some patients presenting with extremely high LDL-C levels. Evinacumab given as a 250-mg subcutaneous injection at baseline and as a 15 mg per kilogram intravenous infusion at week 2 was well tolerated. In a recently reported first-in-human study of healthy human volunteers, evinacumab was also shown to reduce LDL-C and was also well tolerated in a larger number of patients (Dewey, *et al.* 2017 *New Engl J Med*; in press). All nine patients, including the three homozygous null patients lacking LDLR activity, demonstrated clinically meaningful reductions in LDL-C from baseline. Together with recent preclinical studies and human genetic analyses, the results suggest that Angptl3 inhibition can not only lower LDL-C and triglycerides, but also provide

protection from cardiovascular disease. These studies provide real hope for a well-tolerated and impactful treatment for patients suffering from profound familial hypercholesterolemia. Based on the Cholesterol Treatment Trialists' Collaboration (CTTC) analyses (Cholesterol Treatment Trialists (CTT) Collaboration 2010 Lancet 376:1670-81), an absolute decrease of 39 mg/dL in LDL-C corresponds to a 22% relative risk reduction over 4-5 years of statin treatment; recent outcomes data with a PCSK9 antibody support similar risk reductions per unit LDL-C reduction when accounting for shorter treatment duration (Sabatine, *et al*. 2017 *N Engl J Med* 3-17-2017 e-publication; in press). Together with the genetic data on the protective effects of ANGPTL3 mutations on cardiovascular risk (Stitziel, *et al*. 2017 *Journal of the American College of Cardiology* 69(16):2054-2063), the absolute reductions of a 150-200 mg/dL achieved with evinacumab in HoFH patients could have unprecedented benefit for these very high risk patients.

[0148] The mechanism that led to such large reductions in LDL-C is currently under investigation. Evinacumab relieves the normal inhibition, by ANGPTL3, of both lipoprotein lipase (a major regulator of triglycerides and endothelial lipase (a regulator of HDL-C (Shimamura, *et al*. 2007 *Arteriosclerosis, thrombosis, and vascular biology* 27:366-72); thus, the lowering of both triglycerides and HDL-C by evinacumab. The instant results, combined with the results of a recent *in vivo* study (Wang, *et al*. 2015 *J Lipid Res* 56:1296-307), suggest that the effects of evinacumab on LDL-C involve a combination of canonical and noncanonical mechanisms acting upstream of LDL-particle formation. In a mouse model, inactivation of ANGPTL3 with evinacumab did not affect the number of VLDLs secreted by the liver, but qualitatively altered the VLDL particles that were made. Following secretion, VLDLs are rapidly hydrolyzed to form triglyceride-poorer VLDL remnants due to evinacumab-induced up-regulation of lipoprotein lipase, which may increase their clearance through receptors other than LDL receptors. In terms of the modest reductions seen with HDL-C, extensive previous genetic analyses involving endothelial lipase (Voight, *et al*. 2012 *Lancet* 380:572-80), as well as the recent genetic findings with ANGPTL3 protection (Dewey 2017, Stitziel 2017), are consistent with the emerging view that levels of HDL-C do not directly affect cardiovascular risk (Ko, *et al*. 2016 *Journal of the American College of Cardiology* 68:2073-83).

[0149] Patient C and G concomitantly received lomitapide—a microsomal triglyceride transfer protein inhibitor—during the study. These patients showed a 90% and 44% reduction in LDL-C, respectively, 2 weeks after evinacumab intravenous administration, raising the hypothesis of a multiplicative synergy between lomitapide (which affects VLDL production) and evinacumab (which affects the characteristics of secreted VLDL). However, patient D did not receive lomitapide and showed a- 77% reduction in LDL-C at week 4.

[0150] The results reported herein provide proof-of-concept that ANGPTL3 inhibition with evinacumab leads to a substantial additional reduction in LDL-C in patients with HoFH on stable lipid-lowering therapy, including those who have null/null mutations. Evinacumab add-on

therapy allowed normalization of LDL-C concentrations in four HoFH participants in this study. For example, Patient C, a 47-year-old woman, presented LDL-C values above 800 mg per deciliter at age 26. Her lipid profile progressively improved (reaching 150 to 170 mg per deciliter) with the successive introduction of high-dose statins, ezetimibe and lomitapide. LDL-C reached 15 mg per deciliter at week 4, 2 weeks after evinacumab intravenous administration.

Example 3: Inhibition of ANGPTL3 by evinacumab reduced triglycerides (TGs) and LDL-C in subjects presenting with modest elevations in TGs and/or LDL-C

[0151] Elevations in LDL-C and TGs have been linked to increased risk in CHD. Recent discoveries have demonstrated a central role for Angiopoietin like – 3 (ANGPTL3) in lipid metabolism. Loss of function (LoF) of ANGPTL3 in humans has been associated with reductions in TGs, LDL-C, and HDL-C. Evinacumab is a human monoclonal antibody specific for ANGPTL3 that is being developed for treatment of dyslipidemia, including hypertriglyceridemia and hypercholesterolemia.

[0152] Methods: The instant study constituted a phase 1, first-in-human, ascending single-dose, placebo (PBO)-controlled, double-blind study of evinacumab administered subcutaneously (SC) or intravenously (IV) in subjects with elevations of TGs (150≤ TG ≤450 mg/dL) and/or LDL C (≥100 mg/dL). Eighty-three subjects were randomized into the study (9 in PBO SC; 12 in PBO IV; 11 in 75 mg SC: 12 in 150 mg SC, 9 in 250 mg SC, 10 in 5 mg/kg IV, 9 in 10 mg/kg IV, and 11 in 20 mg/kg IV).

[0153] Results: Evinacumab was shown to be well tolerated in this trial. Forty-one (41) subjects reported at least one treatment emergent adverse event (TEAE): 32[± 51.6%] in the evinacumab group vs. 9[± 42.9%] in the PBO group. None were serious, and no subject discontinued due to a TEAE. The most frequent TEAEs were headache (7 [11.3%] vs. 0 [0%]) and increases in ALT/AST [>2X ULN] (5 treated subjects vs 1 PBO subject). There was no dose-related safety trend. Maximum TG reductions were observed on Day 4, with a median % change from baseline of -1.0% to -75.0% across the evinacumab doses and +25.3% for PBO. The mean % changes of LDL-C from baseline on Day 11 were -3.4% to -25.5% across the evinacumab doses and +10.2 for PBO. The duration of TG reduction and LDL was dose-dependent and extended to 64 and 43 days, respectively, after 20 mg/kg IV evinacumab administration. Dose-dependent reductions in HDL-C, VLDL-C, total cholesterol, non-HDL-C, ApoA1, and ApoB were also observed, but there were no apparent effects on Lp(a).

[0154] Administration of evinacumab in healthy subjects with moderately elevated TGs and/or LDL-C was generally well-tolerated. Furthermore, evinacumab induced rapid and substantial reductions in TGs, as well as reductions in LDL-C and HDL-C, recapitulating the observed hypolipoproteinemia in individuals homozygous for ANGPTL3 LOF mutations.

[0155] It is to be understood that, if any prior art publication is referred to herein, such reference does not constitute an admission that the publication forms a part of the common general knowledge in the art, in Australia or any other country.

[0156] In the claims which follow and in the preceding description of the invention, except where the context requires otherwise due to express language or necessary implication, the word "comprise" or variations such as "comprises" or "comprising" is used in an inclusive sense.

i.e. to specify the presence of the stated features but not to preclude the presence or addition of further features in various embodiments of the invention.

We claim:

1.   A method of treating a patient suffering from familial hypercholesterolemia, the method comprising administering to the patient a therapeutically effective amount of a combination of (a) a statin; (b) one lipid lowering agent other than a statin; and (c) an inhibitor of angiopoietin-like protein 3 (ANGPTL3) wherein

    - the inhibitor of angiopoietin-like protein 3 (ANGPTL3) is an antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 and comprises a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8;

    - the statin is selected from the group consisting of atorvastatin, pitavastatin, lovastatin, simvastatin, pravastatin, fluvastatin and rosuvastatin; and

    - the lipid lowering agent other than a statin is an agent that inhibits cholesterol absorption,

and wherein the patient has received or is receiving treatment with a lipid modifying therapy.

2.   The method of claim 1, further comprising administering a therapeutically effective amount of a second lipid-lowering agent other than a statin.

3.   Use of a combination of a statin, one lipid lowering agent other than a statin, and an inhibitor of angiopoietin-like protein 3 (ANGPTL3) in the preparation of a medicament for treating a patient suffering from familial hypercholesterolemia) wherein

    - the inhibitor of angiopoietin-like protein 3 (ANGPTL3) is an antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 and comprises a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8;

    - the statin is selected from the group consisting of atorvastatin, pitavastatin, lovastatin, simvastatin, pravastatin, fluvastatin and rosuvastatin; and

    - the lipid lowering agent other than a statin is an agent that inhibits cholesterol absorption,

and wherein the patient has received or is receiving treatment with a lipid modifying therapy.

4.   Use of an inhibitor of angiopoietin-like protein 3 (ANGPTL3) in the preparation of a medicament for treating a patient suffering from familial hypercholesterolemia wherein

      - the medicament is administered in combination with a statin and one lipid lowering agent other than a statin,

      - the inhibitor of angiopoietin-like protein 3 (ANGPTL3) is an antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 and comprises a heavy chain CDR1 (HCDR1) having the amino acid sequence of SEQ ID NO: 2, a HCDR2 having the amino acid sequence of SEQ ID NO: 3, a HCDR3 having the amino acid sequence of SEQ ID NO: 4, a light chain CDR1 (LCDR1) having the amino acid sequence of SEQ ID NO: 6, a LCDR2 having the amino acid sequence of SEQ ID NO: 7, and a LCDR3 having the amino acid sequence of SEQ ID NO: 8;

      - the statin is selected from the group consisting of atorvastatin, pitavastatin, lovastatin, simvastatin, pravastatin, fluvastatin and rosuvastatin; and

      - the lipid lowering agent other than a statin is an agent that inhibits cholesterol absorption, and

wherein the patient has received or is receiving treatment with a lipid modifying therapy.

5.   The use of claim 3 or 4 wherein the patient is to be administered a therapeutically effective amount of a second lipid-lowering agent other than a statin.

6.   The method of claim 1 or 2, or the use of any one of claims 3-5, wherein the familial hypercholesterolemia is selected from the group consisting of heterozygous familial hypercholesterolemia (HeFH) and homozygous familial hypercholesterolemia (HoFH).

7.   The method or use of any one of claims 1-6, wherein:
   a) the statin is rosuvastatin, administered or to be administered orally once a day at a dose of about 5 mg to about 40 mg; or
   b) the statin is atorvastatin, administered or to be administered orally once a day at a dose of about 10 mg to about 80 mg; or
   c) the agent that inhibits cholesterol absorption is ezetimibe.

8.   The method or use of claim 7, wherein when the agent that inhibits cholesterol absorption is ezetimibe, the ezetimibe is administered or to be administered orally once a day at a dose of about 10 mg.

9.   The method of any one of claims 2 or 6-8, or the use of any one of claims 5 or 6-8, wherein the second lipid lowering agent other than a statin is an agent that inhibits microsomal triglyceride transfer protein (MTTP).

10.   The method or use of claim 9, wherein the agent that inhibits MTTP is lomitapide.

11.   The method or use of claim 10, wherein the lomitapide is administered or to be administered orally once a day at a dose of about 5 mg to about 60 mg.

12.   The method or use of claim 10 or 11, wherein the lomitapide is administered or to be administered orally once a day at a dose of about 20 mg.

13.   The method or use of any one of claims 1-12, wherein the ANGPTL3 antibody is evinacumab.

14.   The method or use of claim 13 wherein evinacumab is administered or to be administered before, during, or after treatment with a statin, ezetimibe, or lomitapide.

15.   The method or use of claim 13 or 14, wherein evinacumab is administered or to be administered:
   a. intravenously at a dose ranging from about 1 mg/kg to about 20 mg/kg of body weight, preferably at a dose of about 15 mg/kg of body weight;
   b. subcutaneously at a dose ranging from about 50 mg to about 750 mg, preferably at a dose ranging from about 250 mg to about 450 mg; or
   c. every week, every two weeks, every 3 weeks, every 4 weeks, every 2 months, every 3 months, or every 4 months.

16.   The method or use of any of one of claims 1-15, wherein the administering, treating or improving results in:
   a. at least a 40% reduction from baseline in at least one lipid parameter; or
   b. at least a 75% reduction from baseline in at least one lipid parameter; optionally in at least a 40% reduction from baseline in LDL-C levels.

17. The method or use of any one of claims 1-16, wherein the antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 comprises the complementarity determining regions (CDRs) of a heavy chain variable (HCVR) having the amino acid

37

sequence of SEQ ID NO: 1 and the CDRs of a light chain variable region (LCVR) of SEQ ID NO: 5.

18.   The method or use of any of claims 1-17, wherein the antibody or antigen-binding fragment thereof that binds specifically to ANGPTL3 comprises a HCVR having the amino acid sequence of SEQ ID NO: 1 and a LCVR having the amino acid sequence of SEQ ID NO: 5.

SEQUENCE LISTING

<110> Regeneron Pharmaceuticals, Inc.

<120> Methods for Treating Patients with
      Familial Hypercholesterolemia

<130> 0431.26PCT

<160> 9

<170> FastSEQ for Windows Version 4.0

<210> 1
<211> 126
<212> PRT
<213> Artificial Sequence

<220>
<223> HCVR

<400> 1
```
Glu Val Gln Leu Val Glu Ser Gly Gly Gly Val Ile Gln Pro Gly Gly
 1               5                   10                  15
Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Asp Asp Tyr
            20                  25                  30
Ala Met Asn Trp Val Arg Gln Gly Pro Gly Lys Gly Leu Glu Trp Val
            35                  40                  45
Ser Ala Ile Ser Gly Asp Gly Gly Ser Thr Tyr Tyr Ala Asp Ser Val
        50                  55                  60
Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Ser Leu Tyr
65                  70                  75                  80
Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Phe Phe Tyr Cys
                85                  90                  95
Ala Lys Asp Leu Arg Asn Thr Ile Phe Gly Val Val Ile Pro Asp Ala
            100                 105                 110
Phe Asp Ile Trp Gly Gln Gly Thr Met Val Thr Val Ser Ser
            115                 120                 125
```

<210> 2
<211> 8
<212> PRT
<213> Artificial Sequence

<220>
<223> HCDR1

<400> 2
```
Gly Phe Thr Phe Asp Asp Tyr Ala
 1               5
```

<210> 3
<211> 8
<212> PRT
<213> Artificial Sequence

<220>
<223> HCDR2

<400> 3
Ile Ser Gly Asp Gly Gly Ser Thr
 1   5

<210> 4
<211> 19
<212> PRT
<213> Artificial Sequence

<220>
<223> HCDR3

<400> 4
Ala Lys Asp Leu Arg Asn Thr Ile Phe Gly Val Val Ile Pro Asp Ala
 1   5   10   15
Phe Asp Ile

<210> 5
<211> 108
<212> PRT
<213> Artificial Sequence

<220>
<223> LCVR

<400> 5
Asp Ile Gln Met Thr Gln Ser Pro Ser Thr Leu Ser Ala Ser Val Gly
 1   5   10   15
Asp Arg Val Thr Ile Thr Cys Arg Ala Ser Gln Ser Ile Arg Ser Trp
                20   25   30
Leu Ala Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile
            35   40   45
Tyr Lys Ala Ser Ser Leu Glu Ser Gly Val Pro Ser Arg Phe Ser Gly
        50   55   60
Ser Gly Ser Gly Thr Glu Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro
65   70   75   80
Asp Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Tyr Asn Ser Tyr Ser Tyr
                85   90   95
Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg
                100   105

<210> 6
<211> 6
<212> PRT
<213> Artificial Sequence

<220>
<223> LCDR1

<400> 6
Gln Ser Ile Arg Ser Trp
 1   5

<210> 7
<211> 3
<212> PRT
<213> Artificial Sequence

<220>
<223> LCDR2

<400> 7
Lys Ala Ser
 1

<210> 8
<211> 9
<212> PRT
<213> Artificial Sequence

<220>
<223> LCDR3

<400> 8
Gln Gln Tyr Asn Ser Tyr Ser Tyr Thr
 1   5

<210> 9
<211> 432
<212> PRT
<213> Artificial Sequence

<220>
<223> hANGPTL3

<400> 9
Met Phe Thr Ile Lys Leu Leu Leu Phe Ile Val Pro Leu Val Ile Ser
 1   5   10   15
Ser Arg Ile Asp Gln Asp Asn Ser Ser Phe Asp Ser Leu Ser Pro Glu
                20   25   30
Pro Lys Ser Arg Phe Ala Met Leu Asp Asp Val Lys Ile Leu Ala Asn
            35   40   45
Gly Leu Leu Gln Leu Gly His Gly Leu Lys Asp Phe Val His Lys Thr
        50   55   60
Lys Gly Gln Ile Asn Asp Ile Phe Gln Lys Leu Asn Ile Phe Asp Gln
65   70   75   80
Ser Phe Tyr Asp Leu Ser Leu Gln Thr Ser Glu Ile Lys Glu Glu Glu
                85   90   95
Lys Glu Leu Arg Arg Thr Thr Tyr Lys Leu Gln Val Lys Asn Glu Glu
                100   105   110
Val Lys Asn Met Ser Leu Glu Leu Asn Ser Lys Leu Glu Ser Leu Leu
            115   120   125
Glu Glu Lys Ile Leu Leu Gln Gln Lys Val Lys Tyr Leu Glu Glu Gln
        130   135   140
Leu Thr Asn Leu Ile Gln Asn Gln Pro Glu Thr Pro Glu His Pro Glu
145   150   155   160
Val Thr Ser Leu Lys Thr Phe Val Glu Lys Gln Asp Asn Ser Ile Lys
                165   170   175
Asp Leu Leu Gln Thr Val Glu Asp Gln Tyr Lys Gln Leu Asn Gln Gln
                180   185   190
His Ser Gln Ile Lys Glu Ile Glu Asn Gln Leu Arg Arg Thr Ser Ile
                195   200   205
Gln Glu Pro Thr Glu Ile Ser Leu Ser Ser Lys Pro Arg Ala Pro Arg
        210   215   220
Thr Thr Pro Phe Leu Gln Leu Asn Glu Ile Arg Asn Val Lys His Asp
225   230   235   240
Gly Ile Pro Ala Glu Cys Thr Thr Ile Tyr Asn Arg Gly Glu His Thr
                245   250   255
Ser Gly Met Tyr Ala Ile Arg Pro Ser Asn Ser Gln Val Phe His Val

```
                    260                      265                      270
Tyr Cys Asp Val Ile Ser Gly Ser Pro Trp Thr Leu Ile Gln His Arg
            275                      280                      285
Ile Asp Gly Ser Gln Asn Phe Asn Glu Thr Trp Glu Asn Tyr Lys Tyr
            290                      295                      300
Gly Phe Gly Arg Leu Asp Gly Glu Phe Trp Leu Gly Leu Glu Lys Ile
305                      310                      315                      320
Tyr Ser Ile Val Lys Gln Ser Asn Tyr Val Leu Arg Ile Glu Leu Glu
                325                      330                      335
Asp Trp Lys Asp Asn Lys His Tyr Ile Glu Tyr Ser Phe Tyr Leu Gly
                340                      345                      350
Asn His Glu Thr Asn Tyr Thr Leu His Leu Val Ala Ile Thr Gly Asn
                355                      360                      365
Val Pro Asn Ala Ile Pro Glu Asn Lys Asp Leu Val Phe Ser Thr Trp
            370                      375                      380
Asp His Lys Ala Lys Gly His Phe Asn Cys Pro Glu Gly Tyr Ser Gly
385                      390                      395                      400
Gly Trp Trp Trp His Asp Glu Cys Gly Glu Asn Asn Leu Asn Gly Lys
                405                      410                      415
Tyr Asn Lys Pro Arg Ala Lys Ser Lys Pro Glu Arg Arg Arg Gly Leu
                420                      425                      430
```