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

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
06 September 2019 (06.09.2019)

WIPO | PCT

(10) International Publication Number
WO 2019/169283 A1

(51) International Patent Classification:
A61P 21/00 (2006.01) *A61K 39/395* (2006.01)

(21) International Application Number:
PCT/US2019/020330

(22) International Filing Date:
01 March 2019 (01.03.2019)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
62/637,017 01 March 2018 (01.03.2018) US

(71) Applicant: **REGENERON PHARMACEUTICALS, INC.** [US/US]; 777 Old Saw Mill River Road, Tarrytown, New York 10591-6707 (US).

(72) Inventors: **DONAHUE, Stephen**; 777 Old Saw Mill River Road, Tarrytown, New York 105914 (US). **PORDY, Robert C.**; 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **HERMAN, Gary**; 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US).

(74) Agent: **KOWALCHYK, Katherine M.** et al.; Merchant & Gould P.C., P.O. BOX 2903, Minneapolis, Minnesota 55402-0903 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:
— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*
— *as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii))*

Published:
— *with international search report (Art. 21(3))*
— *with sequence listing part of description (Rule 5.2(a))*

(54) **Title:** METHODS FOR ALTERING BODY COMPOSITION

(57) **Abstract:** The present invention relates to compositions and methods for altering body composition in a subject, wherein the alteration of body composition is an increase in muscle mass and a reduction of fat mass simultaneously. The present invention also relates to compositions and methods for reducing fat mass in a subject. The compositions and methods also increase muscle volume and lean body mass in the subject. The present invention also relates to compositions that comprise a GDF8 inhibitor and an Activin A inhibitor and the use of such compositions to treat diseases and disorders characterized by increased fat mass, and/or decreased muscle volume.

WO 2019/169283 A1

METHODS FOR ALTERING BODY COMPOSITION

[0001] This application is being filed on March 1, 2019, as a PCT International Patent Application and claims the benefit of priority to U.S. Provisional Application No. 62/637,017, filed on March 1, 2018, the disclosure of which is hereby incorporated by reference in its entirety.

SEQUENCE LISTING

[0002] This application includes a Sequence Listing in electronic format entitled "Sequence-Listing-40848-091WOU1", which was created on 01 March 2019 and which has a size of 288 kilobytes (KB) (295,202 bytes). The contents of txt file "Sequence-Listing-40848-091USU1" are incorporated by reference herein.

FIELD OF THE INVENTION

[0003] The present invention relates to compositions and methods for altering body composition in a subject. The compositions and methods also reduce fat mass in a subject. The compositions and methods also increase muscle volume and/or lean body mass in the subject. More specifically, the invention relates to compositions that comprise a GDF8 inhibitor and an Activin A inhibitor and the use of such compositions to treat diseases and disorders characterized by increased fat mass, and/or decreased muscle volume or decreased lean body mass.

BACKGROUND

[0004] Growth and differentiation factor-8 (GDF8, also known as myostatin), is a secreted ligand belonging to the transforming growth factor-β (TGF-β) superfamily of growth factors. GDF8 plays a central role in the development and maintenance of skeletal muscle, acting as a negative regulator of myogenesis and skeletal muscle mass. Myostatin mutations (including knockout) translate into phenotypes that are predominantly increased muscle mass, but can be made up of variations in muscling (more muscle fibers), muscle fiber composition (greater cross-sectional area of muscle fibers), increased protein/DNA ratio, and other.

[0005] Antibodies to GDF8 and therapeutic methods are disclosed in, *e.g.*, US 8,840,894. Anti-GDF8 antibodies are also mentioned in, *e.g.*, US Patent Nos. 6,096,506; 7,320,789; 7,261,893; 7,807,159; 7,888,486; 7,635,760; 7,632,499; in US Patent Appl. Publ. Nos. 2006/0263354; 2007/0178095; 2008/0299126; 2010/0166764; 2009/0148436;

and International Patent Appl. Publ. Nos. WO2004/037861; WO2007/047112; WO 2010/070094.

[0006] Activins belong to the transforming growth factor-beta (TGF-β) superfamily and exert a broad range of biological effects on cell proliferation, differentiation, metabolism, homeostasis, and apoptosis, as well as immune response and tissue repair. Activin A is a disulfide-linked homodimer (two beta-A chains) that binds to and activates heteromeric complexes of a type I (Act RI-A and Act RI-B) and a type II (Act RII-A and Act RII-B) serine-threonine kinase receptor.

[0007] Antibodies to Activin A and uses thereof are disclosed in, *e.g.*, US 8,309,082; 9,718,881; and International Patent Appl. Publ. No. WO2008/031061.

[0008] Compositions comprising an anti-GDF8 antibody and an anti-Activin A antibody and therapeutic methods are disclosed in, *e.g.*, US 8,871,209.

[0009] Obesity is a global problem for over a third of the world population. In the United States of America, the average obesity rate is over 20%. The costs of obesity-related illness are staggering, amounting to $190.2 billion, roughly 21% of annual medical costs in the U.S. Obesity is an epidemic disease characterized by chronic low-grade inflammation associated with dysfunctional (elevated) fat mass. In Framingham Heart Study participants, abdominal adiposity was associated with incident cardiovascular disease (CVD) after adjustments for clinical risk factors and overall adiposity. Britton JACC 2013 62; 921. Abdominal visceral fat accumulation was positively associated with the progression of coronary noncalcified plaque. Imai Atherosclerosis 2012. Because high fat mass is associated with such serious conditions as congestive heart failure, high blood pressure/hypertension, pulmonary embolism, osteoarthritis, lymphedema, gastro-esophageal reflux disease, chronic renal failure, cancer, fatty-liver disease, and even depression, there remains a need for therapies that reduce total fat and/or android fat mass in subjects.

BRIEF SUMMARY OF THE INVENTION

[0010] In one aspect, the invention is directed to a method for altering body composition in a subject, *i.e.*, increase muscle mass and decreasing fat mass, comprising administering a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor to the subject. In another aspect, the invention is directed to a method for inducing a reduction in fat mass in a subject comprising administering an effective amount of a composition comprising a GDF8 inhibitor and an Activin A inhibitor to the subject.

[0011] In another aspect, the invention is directed to a method for treating a disease or disorder characterized by or associated with increased fat mass, the method comprising administering to a subject in need thereof a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor. In another aspect, the invention is directed to a method for treating a disease or disorder characterized by or associated with increased fat mass, the method comprising administering to a subject in need thereof an effective amount of a composition comprising a GDF8 inhibitor and an Activin A inhibitor. In one aspect, the invention is directed to a method for altering body composition in a subject, *i.e.*, increase muscle mass and decreasing fat mass, comprising administering an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor to the subject, wherein there is an insignificant change in total body mass. Thus, in one aspect of the invention, a subject administered an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor will experience an increase in muscle mass concurrently with a decrease in fat mass, leading to a minimal and/or insignificant change in total body mass.

[0012] In one aspect, the invention is directed to a use of a GDF8 inhibitor and/or an Activin A inhibitor in the preparation of a medicament for achieving a reduction in fat mass in a subject. In another aspect, the invention is directed to a use of a GDF8 inhibitor and/or an Activin A inhibitor in the preparation of a medicament for treating a disease or disorder associated with increased fat mass in a subject.

[0013] In some embodiments, a GDF8 inhibitor is provided for use in a method for treating a disease or disorder characterized by increased fat mass, wherein the method comprises administrating to a subject the GDF8 inhibitor and an Activin A inhibitor.

[0014] In some embodiments, an Activin A inhibitor is provided for use in a method for treating a disease or disorder characterized by increased fat mass, wherein the method comprises administrating to a subject the Activin A inhibitor and a GDF8 inhibitor.

[0015] In some embodiments, a non-therapeutic method is provided for decreasing fat mass in a subject, the method comprising administering to the subject an Activin A inhibitor and a GDF8 inhibitor.

[0016] In one embodiment of a method according to the invention, the effective amount of a GDF8 inhibitor comprises a dosing regimen selected from the group consisting of at least 0.1 mg/kg to about 10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100 mg/kg. In a further embodiment of a method according to the invention, the effective amount of a

GDF8 inhibitor comprises a dosing regimen selected from the group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1 to about 5 mg/kg body weight.

[0017] In another embodiment of a method according to the invention, the effective amount of an Activin A inhibitor comprises a dosing regimen selected from the group consisting of at least 0.1 mg/kg to about 10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100mg/kg. In a further embodiment of a method according to the invention, the effective amount of an Activin A inhibitor comprises a dosing regimen selected from the group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1 to about 5 mg/kg body weight.

[0018] In one embodiment of a method according to the invention, the effective amount of a GDF8 inhibitor is 6 mg/kg body weight and the effective amount of an Activin A inhibitor is 3 mg/kg body weight. In one embodiment of a method according to the invention, the effective amount of a GDF8 inhibitor is 6 mg/kg body weight and the effective amount of an Activin A inhibitor is 10 mg/kg body weight.

[0019] In one embodiment of a method according to the invention, the first composition is formulated for intravenous, subcutaneous, or oral administration. In another embodiment of a method according to the invention, the second composition is formulated for intravenous, subcutaneous, or oral administration. In certain embodiments of a method according to the invention, the first and second compositions are administered concurrently or sequentially to the subject.

[0020] In one embodiment of a method according to the invention, the first and second compositions are combined into a third composition prior to administration. In a further embodiment, the third composition is formulated for intravenous, subcutaneous, or oral administration.

[0021] In one embodiment, a method according to the invention further comprises measuring total fat mass in the subject before administration. In another embodiment, a method according to the invention further comprises measuring total fat mass in the subject after administration, and administering the first and second composition until the subject has a reduction in total fat mass of at least 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more.

[0022] In one embodiment, a method according to the invention further comprises measuring android fat mass in the subject before administration. In another embodiment, a method according to the invention further comprises measuring android fat mass in the

subject after administration, and administering the first and second composition until the subject has a reduction in android fat mass of at least 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more.

[0023] In one embodiment, a method according to the invention further comprises measuring subcutaneous adipose tissue volume in the subject before administration. In another embodiment, a method according to the invention further comprises measuring subcutaneous adipose tissue volume in the subject after administration, and administering the first and second composition until the subject has a reduction in android fat mass of at least 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more.

[0024] In some embodiments, a method is provided comprising administering to a subject in need thereof an effective amount of a GDF8 inhibitor and an effective amount of a Activin A inhibitor, wherein the GDF8 inhibitor and the Activin A inhibitor are co-administered within 48 hours or less, 24 hours or less, 12 hours or less, 6 hours or less, 3 hours or less, or 1 hour or less.

[0025] In some embodiments, a method is provided comprising administering to a subject in need thereof an effective amount of a GDF8 inhibitor and an effective amount of a Activin A inhibitor, wherein the subject exhibits a decrease in total fat mass, android fat mass, and/or subcutaneous adipose tissue volume.

[0026] In some embodiments, a method is provided comprising administering to a subject in need thereof an effective amount of a GDF8 inhibitor and an effective amount of a Activin A inhibitor, wherein the subject exhibits a decrease in total fat mass, android fat mass, and/or subcutaneous adipose tissue volume after 4 weeks or more, or 8 weeks or more, following administration.

[0027] In some embodiments, a method is provided comprising administering to a subject in need thereof an effective amount of a GDF8 inhibitor and an effective amount of a Activin A inhibitor, wherein the subject exhibits a decrease in total fat mass, android fat mass, and/or subcutaneous adipose tissue volume, wherein the subject does not exhibit reduced thigh intramuscular adipose tissue volume.

[0028] In some embodiments, a method is provided comprising administering to a subject in need thereof an effective amount of a GDF8 inhibitor and an effective amount of a Activin A inhibitor, wherein the subject exhibits a decrease in total fat mass, android fat mass, and/or subcutaneous adipose tissue volume, wherein the subject does not exhibit

reduced thigh intramuscular adipose tissue volume after 4 weeks or more, or 8 weeks or more, following administration.

[0029] In some embodiments, a kit is provided comprising a first container containing a effective amount of a GDF8 inhibitor and a second container containing an effective amount of a specific Activin A inhibitor.

[0030] In some embodiments, the GDF8 inhibitor is an isolated antibody or an antigen-binding fragment thereof that specifically binds to GDF8.

[0031] In one embodiment of a method according to the invention, the GDF8 inhibitor is an antibody or an antigen-binding fragment thereof that specifically binds to GDF8. In another embodiment, the antibody or antigen-binding fragment that specifically binds GDF8 comprises the heavy chain complementarity determining regions (HCDRs) of a heavy chain variable region (HCVR) comprising SEQ ID NO:360, and the light chain complementarity determining regions (LCDRs) of a light chain variable region (LCVR) comprising SEQ ID NO:368. In still another embodiment, the antibody or antigen-binding fragment that specifically binds GDF8 comprises three HCDRs comprising SEQ ID NO:362, SEQ ID NO:364, and SEQ ID NO:366, and three LCDRs comprising SEQ ID NO:370, SEQ ID NO:372, and SEQ ID NO:374.

[0032] In some embodiments, the Activin A inhibitor is an isolated antibody or an antigen-binding fragment thereof that specifically binds to Activin A.

[0033] In one embodiment of a method according to the invention, the Activin A inhibitor is an antibody or antigen-binding fragment thereof that specifically binds Activin A. In another embodiment, the antibody or antigen-binding fragment that specifically binds Activin A comprises the heavy chain complementarity determining regions (HCDRs) of a heavy chain variable region (HCVR) comprising SEQ ID NO:553, and the light chain complementarity determining regions (LCDRs) of a light chain variable region (LCVR) comprising SEQ ID NO:537. In still another embodiment, the antibody or antigen-binding fragment that specifically binds Activin A comprises three HCDRs comprising SEQ ID NO:555, SEQ ID NO:557, and SEQ ID NO:559, and three LCDRs comprising SEQ ID NO:539, SEQ ID NO:541, and SEQ ID NO:543.

[0034] In one embodiment of a method according to the invention, the effective dose of the Activin A inhibitor is selected from the group comprising between 100% to 200% of the effective dose of the GDF8 inhibitor, between 100% and 250% of the effective dose of the GDF8 inhibitor, between 100% and 300% of the effective dose of the GDF8 inhibitor, and between 100% and 400% by weight of the effective dose of the GDF8 inhibitor.

[0035] In another embodiment of a method according to the invention, the weight ratio of the effective dose of the Activin A inhibitor to effective dose of the GDF8 inhibitor is from 10:1 to 1:10, 8:1 to 1:8, 6:1 to 1:6, 3:1 to 1:3, or about 2:1 to 1:2. In another embodiment of a method according to the invention, the weight ratio of the effective dose of the Activin A inhibitor is about 1.5 to 2.0 times as large by weight as the amount the GDF8 inhibitor.

[0036] In one embodiment of a method according to the invention, the GDF8 inhibitor is a bispecific antibody or antigen-binding fragment thereof that specifically binds GDF8 and also specifically binds Activin A. In another embodiment, the Activin A inhibitor is a bispecific antibody or antigen-binding fragment thereof that specifically binds Activin A and also specifically binds GDF8.

[0037] In one embodiment of a method according to the invention, the reduction of fat mass in the subject is a reduction in total fat mass as measured by DXA (Dual-energy X-ray absorptiometry). In another embodiment of a method according to the invention, the reduction of fat mass in the subject is a reduction in android fat mass as measured by DXA (Dual-energy X-ray absorptiometry).

[0038] In one embodiment of a method according to the invention, the reduction of fat mass in the subject is a reduction in subcutaneous adipose tissue volume as measured by MRI (Magnetic Resonance Imaging).

[0039] In one embodiment of a method according to the invention, the subject experiences an increase in muscle volume. The muscle volume may be thigh muscle tissue volume, for example, as measured by MRI. In some embodiments, the muscle volume may be thigh muscle tissue volume, for example, as measured by MRI. In some embodiments, the thigh muscle volume may be thigh muscle tissue volume including intramuscular adipose tissue and large vessels, or thigh muscle tissue volume excluding intramuscular adipose tissue and large vessels, for example, as measured by MRI.

[0040] In one embodiment of a method according to the invention, the subject experiences an increase in total lean mass. The total lean mass may be measured by DXA (dual x-ray absorptiometry.

[0041] In one embodiment of a method according to the invention, the subject experiences an increase in appendicular lean body mass. The appendicular lean body mass may be measured by DXA, and, for example, calculated by aLBM equation.

[0042] In one embodiment of a method according to the invention, the subject experiences a decrease in total fat mass, for example, as measured by DXA.

[0043] In one embodiment of a method according to the invention, the subject experiences a decrease in android fat mass, for example, as measured by DXA.

[0044] In one embodiment of a method according to the invention, the subject experiences a decrease in subcutaneous adipose tissue volume, for example, as measured by DXA.

[0045] In one embodiment of a method according to the invention, the subject experiences a decrease in sum of fat mass of arms and legs, for example, as measured by DXA.

[0046] In one embodiment of the invention, the subject does not exhibit a decrease in thigh intramuscular adipose tissue volume, for example, as measured by MRI.

[0047] In one embodiment of the invention, the subject does not exhibit a decrease in total bone mineral density (BMD) mass, for example, as measured by DXA.

[0048] In one embodiment of the invention, the subject does not exhibit a decrease in total bone mineral content (BMC) mass, for example, as measured by DXA.

[0049] In one embodiment of the invention, the subject exhibits an increase in total bone mineral content (BMC) mass, for example, as measured by DXA.

[0050] In another embodiment of a method according to the invention, the subject does not have a muscle wasting condition or disease.

[0051] In some embodiments, a kit is provided for use in altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass, the kit comprising a first container comprising a composition comprising an effective amount of a GDF8 inhibitor and a second container comprising a second composition comprising an effective amount of an Activin A inhibitor.

[0052] In some embodiments, a GDF8 inhibitor is provided for use in manufacture of a first composition for use as a medicament in a kit for altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass, the kit further comprising a second composition comprising an Activin A inhibitor.

[0053] In some embodiments, an Activin A inhibitor is provided for use in manufacture of a first composition for use as a medicament in a kit for altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass, the kit further comprising a second composition comprising a GDF8 inhibitor.

[0054] In some embodiments, a GDF8 inhibitor is provided for use in manufacture of a first composition for use in a kit for altering body composition, decreasing fat mass, or increasing lean mass in a subject, the kit further comprising a second composition comprising an Activin A inhibitor.

[0055] In some embodiments, an Activin A inhibitor is provided for use in manufacture of a first composition for use in a kit for altering body composition, decreasing fat mass, or increasing lean mass in a subject, the kit further comprising a second composition comprising a GDF8 inhibitor.

[0056] In some embodiments, a first composition comprising a GDF8 inhibitor is provided for use in altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass or decreased lean mass in a subject, wherein the subject has received a second composition comprising an Activin A inhibitor.

[0057] In some embodiments, a first composition comprising an Activin A inhibitor is provided for use in altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass or decreased lean mass in a subject, wherein the subject has received a second composition comprising a GDF8 inhibitor.

[0058] In some embodiments, a first composition comprising a GDF8 inhibitor is provided for use in method for altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass or decreased lean mass, the method further comprising administering a second composition comprising an Activin A inhibitor.

[0059] In some embodiments, a first composition comprising an Activin A inhibitor is provided for use in method for altering body composition, decreasing fat mass, increasing lean mass, or treating a disease or disorder characterized by or associated with increased fat mass or decreased lean mass, the method further comprising administering a second composition comprising an GDF8 inhibitor.

[0060] In some embodiments, a composition is provided comprising an Activin A inhibitor and a GDF8 inhibitor for use in altering body composition, decreasing fat mass, increasing lean mass, or treating or preventing a disease or disorder characterized by or associated with increased fat mass or decreased lean mass.

[0061] Other embodiments of the present invention will become apparent from a review of the ensuing detailed description.

BRIEF DESCRIPTION OF THE FIGURES

[0062] Figure 1 shows a bar graph depicting results of clinical studies in sarcopenia patients 70 years of age or older receiving anti-GDF8 antibody REGN1033 alone after 12 weeks as the percent change from baseline of total lean mass in LS mean and SE compared to placebo. Patients receiving REGN1033 exhibited significantly increased total lean body mass at 12 weeks at each of three dosing regimens when compared to placebo (n=65). Patients receiving 100 mg anti-GDF8 antibody REGN1033 Q4W S.C. exhibited a difference vs. placebo of 1.66% total lean mass (n=62, P=0.0077). Patients receiving 300 mg anti-GDF8 Q4W SC exhibited a difference vs. placebo of 1.78% total lean mass (n=64, P=0.0043). Patients receiving 300 mg Q2W SC exhibited a difference vs. placebo of 2.29% total lean mass (n=59, P=0.0004).

[0063] Figure 2A shows Table 1 with the ascending dose panels used in the study of 48 healthy post-menopausal women according to Example 2. A single intravenous dose of anti-GDF8 antibody REGN1033 and/or anti-Activin A antibody REGN2477 was utilized. In the primary analyses, the placebo and high dose combination groups were pooled across panels, yielding 12 subjects on placebo and 12 on the high dose combination, as shown by boxed regions in the table.

[0064] Figure 2B shows a bar graph depicting thigh muscle volume (measured via MRI) % change at week 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women. A significant increase in thigh muscle volume % change from placebo was exhibited by anti-GDF8 (6 mg/kg), anti-GDF8 (6 mg/kg)+ mid dose anti-Activin A (3 mg/kg), and anti-GDF8 (6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups. (*nominal $p < 0.5$ vs. placebo, **** nominal $p < 0.0001$ vs. placebo).

[0065] Figure 2C shows a bar graph depicting total fat mass (measured via DXA) % change at week 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women. The numbers show changes from placebo. A significant decrease in total fat mass % change was exhibited by the anti-GDF8 (6 mg/kg) + high dose anti-Activin A (10 mg/kg) group. (*nominal $p < 0.05$ vs. placebo).

[0066] Figure 3 shows a line graph depicting LS mean (SE) percent change from baseline in thigh muscle volume by MRI at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women. At 4 weeks after single I.V. dose, a significant increase in thigh

muscle volume (measured by MRI excluding intramuscular adipose tissue) % change from placebo was exhibited by each of anti-GDF8 (6 mg/kg), anti-GDF8 (6 mg/kg)+ low dose anti-Activin A (1 mg/kg), anti-GDF8 (6 mg/kg)+ mid dose anti-Activin A (3 mg/kg), and anti-GDF8 (6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups. At 8 weeks after single I.V. dose, a significant increase in thigh muscle volume % change from placebo was exhibited in anti-GDF8 (6 mg/kg)+ mid dose anti-Activin A (3 mg/kg), and anti-GDF8 (6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups. (*nominal $p<0.05$, **nominal $p<0.001$). N values for each group are shown in Figure 2A.

[0067] Figure 4 show line graphs depicting individual data for percent change (from baseline) in thigh muscle volume by MRI at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women as % change from baseline in placebo, anti-GDF8 (6 mg/kg), high dose anti-Activin A (10 mg/kg), anti-GDF8(6 mg/kg) + low dose anti-Activin A(1 mg/kg), anti-GDF8(6 mg/kg) + mid dose anti-Activin A(3 mg/kg), and anti-GDF8(6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups for multiple individuals. Increases in thigh muscle volume were consistently observed in individual subjects following treatment with REGN2477 + REGN1033 in combination. Within each treatment group, different lines indicate different individuals.

[0068] Figure 5 shows a line graph depicting appendicular lean (body) mass (*i.e.*, sum of lean tissue in the arms and legs) at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women as % percent change from baseline LS (least-squares) mean (SE) in six groups. N values for each group are shown in Figure 2A. After 4 weeks and 8 weeks, each of the three combination dose groups, including anti-GDF8(6 mg/kg) + low dose anti-Activin A(1 mg/kg), anti-GDF8(6 mg/kg) + mid dose anti-Activin A(3 mg/kg), and anti-GDF8(6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups, exhibited significantly increased % change LS mean difference in appendicular lean mass compared to placebo (* nominal $p<0.05$, ** nominal $p<0.001$). N values for each group are shown in Figure 2A.

[0069] Figure 6 shows a line graph depicting mean (SE = standard errors) total fat mass percent (as measured by DXA) change at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in placebo, anti-GDF8 (6 mg/kg), high dose anti-Activin A (10 mg/kg), anti-GDF8(6 mg/kg) + low dose anti-Activin A(1 mg/kg), anti-GDF8(6 mg/kg) +

mid dose anti-Activin A(3 mg/kg), and anti-GDF8(6 mg/kg) + high dose anti-Activin A (10 mg/kg) groups. N values for each group are shown in Figure 2A. The high dose group, (anti-GDF8(6 mg/kg) + high dose anti-Activin A (10 mg/kg), exhibited a significant reduction in total fat mass as percent change LS Mean difference compared to placebo at week 4 and week 8 (*nominal $p < 0.05$). Blockade of both Activin A and GDF8 led to reductions in total fat mass, as assessed by DXA.

[0070] Figure 7 shows a line graph depicting mean (SE) percent change in android fat mass at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8(6 mg/kg), high dose (10 mg/kg) anti-Activin A, anti-GDF8(6 mg/kg) + low dose (1 mg/kg) anti-Activin A, anti-GDF8(6 mg/kg) + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 (6 mg/kg)+ high dose (10 mg/kg) anti-Activin A groups. The high dose REGN1033 + REGN2477 group exhibited significantly reduced percent change LS Mean difference in android fat mass by DXA compared to placebo at week 4 and week 8 (*nominal $p < 0.05$). N values for each group are shown in Figure 2A. Blockade of both Activin A and GDF8 was also associated with decreases in android fat mass, as assessed by DXA.

[0071] Figure 8 shows a line graph depicting LS mean percent change with SE in thigh muscle volume (excluding intramuscular adipose tissue and large vessels) at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8(6 mg/kg), high dose (10 mg/kg) anti-Activin A, anti-GDF8 (6 mg/kg)+ low dose (1 mg/kg) anti-Activin A, anti-GDF8(6 mg/kg) + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8(6 mg/kg) + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. Compared with placebo, REGN2477 +REGN1033 medium and high groups exhibited significantly increased mean % change in thigh muscle volume at 4 weeks and 8 weeks. (*nominal $p < 0.05$, **nominal $p < 0.001$).

[0072] Figure 9 shows a line graph depicting LS mean percent change with SE in total lean mass at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women for six groups including placebo, anti-GDF8(6 mg/kg), high dose (10 mg/kg) anti-Activin A, anti-GDF8 (6 mg/kg)+ low dose (1 mg/kg) anti-Activin A, anti-GDF8 (6 mg/kg)+ mid dose (3 mg/kg) anti-Activin A, and anti-GDF8(6 mg/kg) + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A.

Compared with placebo, REGN2477 +REGN1033 medium and high dose groups exhibited significantly increased % change in total lean mass compared to placebo at 4 weeks and 8 weeks. (*nominal $p < 0.05$).

[0073] Figure 10 shows a line graph depicting LS mean percent change with SE in appendicular lean body mass (calculated via aLBM equation), in kg, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. REGN2477 + REGN1033 treatment resulted in significantly increased % change in appendicular lean body mass calculated by aLBM equation)(kg) at each low, medium and high dose groups at 4 weeks and 8 weeks compared with placebo (* $p<0.05$, ** $p < 0.001$).

[0074] Figure 11 shows a line graph depicting LS mean percent change with SE in total fat mass, in kg, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8 (6 mg/kg), high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8(6 mg/kg) + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 (6 mg/kg) + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The high dose combination REGN2477 + REGN1033 treatment group exhibited significantly decreased % total fat mass: -3.92% (high dose group) compared with placebo -0.65% at 8 weeks. (*nominal $p<0.05$).

[0075] Figure 12 shows a line graph depicting LS mean percent change with SE in thigh muscle volume, in cm^3, (including intramuscular adipose tissue and large vessels) at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The high and medium dose REGN2477 + REGN1033 treatment groups exhibited significantly increased % change in thigh muscle volume, including intramuscular adipose tissue and large vessels in medium and high treatment groups at 4 weeks and 8 weeks compared to placebo (*nominal $p<0.05$, **nominal $p < 0.001$).

[0076] Figure 13 shows a line graph depicting LS mean percent change with SE in appendicular lean mass (sum of lean mass of arms and legs), in kg, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. Appendicular lean mass was significantly increased as mean percent change from placebo in each REGN2477 + REGN1033 low, medium and high treatment groups at 4 and 8 weeks (*nominal $p<0.05$, *nominal* $p<0.001$).

[0077] Figure 14 shows a line graph depicting LS mean percent change with SE in android fat mass, in kg, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The high dose REGN1033 + REGN2477 group exhibited significantly reduced % change in android fat mass by DXA compared to placebo at week 4 and week 8 (*nominal $p<0.05$).

[0078] Figure 15 shows a line graph depicting LS mean percent change with SE in thigh intramuscular adipose tissue volume, in cm^3, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The high dose REGN1033 + REGN2477 group exhibited increased thigh intramuscular adipose tissue volume as mean % change compared to placebo at 8 weeks. (*nominal $p < 0.05$).

[0079] Figure 16 shows a line graph depicting LS mean percent change with SE in sum of intramuscular and perimuscular adipose tissue (IMAT) at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose

(10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The combination REGN1033 + REGN2477 treatment groups were not significantly different than placebo in LS mean percent change with SE in sum of intramuscular and perimuscular adipose tissue (IMAT) at weeks 4 and 8.

[0080] Figure 17 shows a line graph depicting LS mean percent change with SE in subcutaneous adipose tissue volume, in cm^3, at weeks 0, 4, and 8 after a single I.V. dose of anti-Activin A antibody REGN2477 and/or anti-GDF8 antibody REGN1033 in healthy post-menopausal women in six groups including placebo, anti-GDF8, high dose (10 mg/kg) anti-Activin A, anti-GDF8 + low dose (1 mg/kg) anti-Activin A, anti-GDF8 + mid dose (3 mg/kg) anti-Activin A, and anti-GDF8 + high dose (10 mg/kg) anti-Activin A groups. N values for each group are shown in Figure 2A. The REGN1033 + REGN2477 low and medium treatment groups exhibited significantly decreased % change in subcutaneous adipose tissue volume compared to placebo at 8 weeks (* nominal $p<0.05$).

DETAILED DESCRIPTION

[0081] Before the present invention is described, it is to be understood that this invention is not limited to particular compositions, methods, and experimental conditions described, as such compositions, 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. 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.

[0082] As used in this specification and the appended claims, the singular forms “a”, “an”, and “the” include plural references unless the context clearly dictates otherwise. Thus, for example, a reference to “a method” includes one or more methods, and/or steps of the type described herein and/or which will become apparent to those persons skilled in the art upon reading this disclosure.

[0083] 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.).

[0084] 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 in their entirety.

Antibodies and Antigen-Binding Fragments of Antibodies

[0085] 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 antibodies (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.

[0086] Antibodies may be referred to herein according to the following nomenclature: Fc prefix (*e.g.* "H1M," "H2aM," "H4H"), followed by a numerical identifier (*e.g.* "10446"), followed by a "P," "P2" or "N" suffix. Thus, according to this nomenclature, an antibody may be referred to herein as, *e.g.*, "H4H10446P2". The H1M, H2M and H4H prefixes on the antibody designations used herein indicate the particular Fc region isotype of the antibody. For example, an "H2aM" antibody has a mouse IgG2a Fc, whereas an "H4H" antibody has a human IgG4 Fc. As will be appreciated by a person of ordinary skill in the art, an antibody having a particular Fc isotype can be converted to an antibody with a different Fc isotype (*e.g.*, an antibody with a mouse IgG2a Fc can be converted to an antibody with a human IgG4, etc.), but in any event, the variable domains (including the CDRs) will remain the same, and the binding properties are expected to be identical or substantially similar regardless of the nature of the Fc domain.

[0087] 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.

[0088] 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.

[0089] 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 that 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.

[0090] 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 used in 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 used in 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)).

[0091] Full antibody molecules and antigen-binding fragments may be monospecific or multispecific (*e.g.*, bispecific). A multispecific antibody or 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 may be adapted for use in the context of an antibody or antigen-binding fragment described herein using routine techniques available in the art.

[0092] The antibodies used in the compositions and methods of the invention may function through complement-dependent cytotoxicity (CDC) or antibody-dependent cell-mediated cytotoxicity (ADCC). "Complement-dependent cytotoxicity" (CDC) refers to lysis of antigen-expressing cells by an antibody of the invention in the presence of complement. "Antibody-dependent cell-mediated cytotoxicity" (ADCC) refers to a cell-mediated reaction in which nonspecific cytotoxic cells that express Fc receptors (FcRs) (*e.g.*, Natural Killer (NK) cells, neutrophils, and macrophages) recognize bound antibody on a target cell and thereby lead to lysis of the target cell. CDC and ADCC can be measured using assays that are well known and available in the art. (*See, e.g.*, U.S. Pat. Nos. 5,500,362 and 5,821,337, and Clynes *et al.*, *Proc. Natl. Acad. Sci. (USA) 95*:652-656 (1998)).

[0093] The antibodies described herein may comprise or consist of human antibodies and/or recombinant human antibodies, or fragments thereof. The term "human antibody", as used herein, includes antibodies having variable and constant regions derived from human germline immunoglobulin sequences. Human antibodies 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.

[0094] The antibody molecules described herein may comprise or consist of recombinant human antibodies or antigen-binding fragments thereof. 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, antibodies isolated from a recombinant, combinatorial human antibody library, 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*. Recombinant antibodies may be used in an additional embodiment of the compositions and methods of the invention.

[0095] 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, tissue or cell in which the antibody naturally exists or is naturally produced is an "isolated antibody" for purposes of the invention. An isolated antibody also includes an antibody *in situ* within a recombinant cell, as well as an antibody that has 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. Isolated antibodies may be used in an additional embodiment of the compositions and methods of the invention.

[0096] A protein or polypeptide is "substantially pure," "substantially homogeneous" or "substantially purified" when at least about 60 to 75% of a sample exhibits a single species of polypeptide. The polypeptide or protein may be monomeric or multimeric. A substantially pure polypeptide or protein will typically comprise about 50%, 60, 70%, 80% or 90% w/w of a protein sample, usually about 95%, and preferably over 99% pure. Protein purity or homogeneity may be indicated by a number of means well known in the art, such as polyacrylamide gel electrophoresis of a protein sample, followed by visualizing a single polypeptide band upon staining the gel with a stain well known in the art. For certain purposes, higher resolution may be provided by using HPLC or other means well known in the art for purification.

[0097] The term "polypeptide analog or variant" as used herein refers to a polypeptide that is comprised of a segment of at least 25 amino acids that has substantial identity to a portion of an amino acid sequence and that has at least one of the following properties: (1) specific binding to GDF8 under suitable binding conditions, or (2) ability to block the biological activity of GDF8. Typically, polypeptide analogs or variants comprise a conservative amino acid substitution (or insertion or deletion) with respect to the naturally occurring sequence. Analogs typically are at least 20 amino acids long, at least 50, 60, 70, 80, 90, 100, 150 or 200 amino acids long or longer, and can often be as long as a full-length naturally-occurring polypeptide.

[0098] Preferred amino acid substitutions are those which: (1) reduce susceptibility to proteolysis, (2) reduce susceptibility to oxidation, (3) alter binding affinity for forming protein complexes, (4) alter binding affinities, and (4) confer or modify other physicochemical or functional properties of such analogs. Analogs can include various mutations of a sequence other than the naturally-occurring peptide sequence. For example, single or multiple amino acid substitutions (preferably conservative amino acid substitutions) may be made in the naturally occurring sequence (preferably in the portion of the polypeptide outside the domain(s) forming intermolecular contacts. A conservative amino acid substitution should not substantially change the structural characteristics of the parent sequence (*e.g.*, a replacement amino acid should not tend to break a helix that occurs in the parent sequence, or disrupt other types of secondary structure that characterizes the parent sequence). Examples of art-recognized polypeptide secondary and tertiary structures are described in Proteins, Structures and Molecular Principles (Creighton 1984 W. H. Freeman and Company, New York; Introduction to Protein

Structure (Branden & Tooze, eds., 1991, Garland Publishing, NY); and Thornton et at. 1991 Nature 354:105, which are each incorporated herein by reference.

[0099] Non-peptide analogs are commonly used in the pharmaceutical industry as drugs with properties analogous to those of the template peptide. These types of non-peptide compound are termed "peptide mimetics" or "peptidomimetics" (see, for example, Fauchere (1986) J. Adv. Drug Res. 15:29; and Evans et al. (1987) J. Med. Chem. 30:1229, which are incorporated herein by reference. Systematic substitution of one or more amino acids of a consensus sequence with a D-amino acid of the same type (*e.g.*, D-lysine in place of L-lysine) may also be used to generate more stable peptides. In addition, constrained peptides comprising a consensus sequence or a substantially identical consensus sequence variation may be generated by methods known in the art (Rizo et al. (1992) Ann. Rev. Biochem. 61:387, incorporated herein by reference), for example, by adding internal cysteine residues capable of forming intramolecular disulfide bridges which cyclize the peptide.

[00100] As applied to polypeptides, the term "substantial identity" or "substantially identical" means that two peptide sequences, when optimally aligned, such as by the programs GAP or BESTFIT using default gap weights, share at least about 80% sequence identity, at least about 90%, at least about 95%, at least about 98% or at least about 99% sequence identity. Preferably, residue positions that are not identical differ by conservative amino acid substitutions. A "conservative amino acid substitution" is one in which an amino acid residue is substituted by another amino acid residue having a side chain (R group) with similar chemical properties (*e.g.*, charge or hydrophobicity). In general, a conservative amino acid substitution will not substantially change the functional properties of a protein. In cases where two or more amino acid sequences differ from each other by conservative substitutions, the percent sequence identity or degree of similarity may be adjusted upwards to correct for the conservative nature of the substitution. Means for making this adjustment are well-known to those of skill in the art. See, *e.g.*, Pearson (1994) Methods Mol. Biol. 24:307-331, herein incorporated by reference. Examples of groups of amino acids that have side chains with similar chemical properties include 1) aliphatic side chains: glycine, alanine, valine, leucine and isoleucine; 2) aliphatic-hydroxyl side chains: serine and threonine; 3) amide-containing side chains: asparagine and glutamine; 4) aromatic side chains: phenylalanine, tyrosine, and tryptophan; 5) basic side chains: lysine, arginine, and histidine; and 6) sulfur-containing side chains are cysteine and methionine. Preferred conservative amino acids substitution groups are: valine-

leucine-isoleucine, phenylalanine-tyrosine, lysine-arginine, alanine-valine, glutamate-aspartate, and asparagine-glutamine. Alternatively, a conservative replacement is any change having a positive value in the PAM250 log-likelihood matrix disclosed in Gonnet et al. (1992) Science 256:1443-45, herein incorporated by reference. A "moderately conservative" replacement is any change having a nonnegative value in the PAM250 log-likelihood matrix.

[00101] Sequence similarity for polypeptides, which is also referred to as sequence identity, is typically measured using sequence analysis software. Protein analysis software matches similar sequences using measures of similarity assigned to various substitutions, deletions and other modifications, including conservative amino acid substitutions. For instance, GCG contains programs such as "Gap" and "Bestfit" which can be used with default parameters to determine sequence homology or sequence identity between closely related polypeptides, such as homologous polypeptides from different species of organisms or between a wild type protein and a mutein thereof. See, *e.g.*, GCG Version 6.1. Polypeptide sequences also can be compared using FASTA using default or recommended parameters, a program in GCG Version 6.1. FASTA (*e.g.*, FASTA2 and FASTA3) provides alignments and percent sequence identity of the regions of the best overlap between the query and search sequences (Pearson (2000), *supra*). Another preferred algorithm when comparing a sequence of the invention to a database containing a large number of sequences from different organisms is the computer program BLAST, especially blastp or tblastn, using default parameters. See, *e.g.*, Altschul et al. (1990) J. Mol. Biol. 215:403-410 and Altschul et al. (1997) Nucleic Acids Res. 25:3389 402, each of which is herein incorporated by reference.

[00102] The length of polypeptide sequences compared for homology will generally be at least about 16 amino acid residues, at least about 20 residues, at least about 24 residues, at least about 28 residues, or at least about 35 residues. When searching a database containing sequences from a large number of different organisms, it is preferable to compare amino acid sequences.

[00103] In some embodiments, the invention relates to a method for altering the body composition of a subject comprising administering a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor to the subject.

[00104] In some embodiments, the invention relates to a method for inducing a reduction in fat mass in a subject comprising administering a first composition comprising an

effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor to the subject.

[00105] In some embodiments, the invention relates to a method for inducing an increase in muscle mass in a subject, the method comprising administering to a subject in need thereof a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor.

[00106] In some embodiments, the invention relates to a method for altering the body composition of a subject comprising administering a composition comprising an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor to the subject.

[00107] In some embodiments, the invention relates to a method for inducing a reduction in fat mass in a subject comprising administering a composition comprising an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor to the subject.

[00108] In some embodiments, the invention relates to a method for inducing an increase in muscle mass in a subject, the method comprising administering to a subject in need thereof a composition comprising an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor.

[00109] In some embodiments, the subject is a human subject. The human subject may be an adult human subject. The subject may be a male or female subject. The subject may be a healthy subject. The subject may be suffering from unwanted fat mass. The subject may be suffering from, or at risk of suffering from, a disease and disorder characterized by increased fat mass, and/or decreased muscle volume or decreased lean body mass. The subject may be a post-menopausal female subject. The subject may be a male subject or female subject 40 years of age or older, 50 years of age or older, 60 years of age or older, or 70 years of age or older.

[00110] In some embodiments, the invention relates to a method comprising administering a composition comprising a GDF8 inhibitor and a composition comprising an Activin A inhibitor to a subject in need thereof for treating or preventing diseases and disorders characterized by increased fat mass, and/or decreased muscle volume or decreased lean body mass.

[00111] In some embodiments according to the invention, the subject has at least one disease or disorder that may be associated with increased fat mass. In some embodiments, the disease or disorder may be selected from the group consisting of obesity, metabolic

syndromes, nutritional disorders, high cholesterol, dyslipidemia, cardiovascular disease, cellulitis, cancer (including of the colon, esophagus, kidney, pancreas, gallbladder, breast, or endometrium), polycystic ovarian syndrome, gout, gallbladder disease, sleep apnea, respiratory disorder, asthma, osteoarthritis, cataract, congestive heart failure, enlarged heart, high blood pressure/hypertension, pulmonary embolism, lymphedema, gastro-esophageal reflux disease, hernia, chronic renal failure, urinary incontinence, connective tissue diseases, and fatty-liver disease. In another embodiment, the disease or disorder may be sarcopenia.

GDF8 inhibitors

[00112] The present invention includes methods for altering body composition, inducing a reduction in fat mass, and increasing lean mass in a subject, and methods for treating a disease or disorder characterized by increased fat mass in a subject, comprising administering a composition comprising an effective amount of a GDF8 inhibitor to the subject.

[00113] The term "GDF8" (also referred to as "growth and differentiation factor-8" and "myostatin") means the protein having the amino acid sequence of SEQ ID NO:340 (mature protein). According to the present invention, GDF8-specific binding proteins specifically bind GDF8 but do not bind other ActRIIB ligands such as GDF3, BMP2, BMP4, BMP7, BMP9, BMP10, GDF11, Activin A, Activin B, Activin AB, Nodal, *etc.*

[00114] As used herein, a "GDF8 inhibitor" is any agent that binds to or interacts with human GDF8 and interferes with or inhibits the normal biological function of GDF8 *in vitro* or *in vivo*. Non-limiting examples of categories of GDF8 inhibitors include small molecule GDF8 antagonists, nucleic acid-based inhibitors of GDF8 expression or activity (*e.g.*, siRNA or antisense), peptide-based molecules that specifically interact with GDF8 (*e.g.*, peptibodies), receptor molecules that specifically interact with GDF8, GDF8-binding scaffold molecules, proteins comprising a ligand-binding portion of a receptor that specifically binds GDF8, and anti-GDF8 aptamers or portions thereof. In a preferred embodiment, a GDF8 inhibitor that can be used in the context of the present invention is an anti-GDF8 antibody or antigen-binding fragment thereof that specifically binds human GDF8. Anti-GDF8 antibodies include neutralizing and/or blocking antibodies. The inhibition caused by anti-GDF8 neutralizing and/or blocking antibodies need not be complete, as long as it is detectable using appropriate assays.

[00115] As used herein, the expression "anti-GDF8 antibody" also includes multispecific antigen-binding molecules (*e.g.*, bispecific antibodies), wherein at least one binding

domain (*e.g.*, "binding arm") of the multispecific antigen-binding molecule specifically binds GDF8.

[00116] Exemplary anti-GDF8 antibodies that can be used in the compositions and methods of the invention include, *e.g.*, the fully-human anti-GDF8 antibody H4H1657N2, also known as REGN1033, (*e.g.*, an anti-GDF8 antibody comprising the heavy and light chain variable regions having amino acid sequences SEQ ID NO: 360 and SEQ ID NO: 368, respectively, as set forth in US Patent No. 8,840,894). Other GDF8 antagonists that can be used in the compositions and methods of the invention include anti-GDF8 antibodies (*e.g.*, the antibody designated 2_112_1 , *e.g.*, having ATCC deposit designation PTA-6574, or *e.g.*, 2_112_K, *e.g.*, having HCVR/LCVR amino acid sequences SEQ ID NOs: 620 and 621) as set forth in US 2006/0263354 and US Patent No. 7,807,159; anti-GDF8 antibodies (*e.g.*, 12A5-5, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO: 622 and 623) as set forth in US Patent No. 8,999,343 and US Publication No. 2013/0209489; anti-GDF8 antibodies (*e.g.*, 10B3H8L5, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO:624 and 625, and 10B3H8L5-Fc-disabled) as set forth in US Publication No. 2013/0142788; anti-GDF8 antibodies (*e.g.*, stamulumab/MYO-29, *e.g.*, having HCVR/LCVR amino acid sequences of SEQ ID NOs: 626 and 627) as set forth in US Patent Nos. 8,940,874 and 7,261,893; anti-GDF8 antibodies (*e.g.*, RK22/PF-0625616, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO: 628 and 629) as set forth in US Patent No. 8,415,459; anti-GDF8 antibodies (*e.g.*, JA-16, e.g., having CDRs of HCVR amino acid sequence of SEQ ID NO: 630) as set forth in US Patent No. 7,731,961; anti-GDF8 antibodies (*e.g.*, RK35, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO: 631 and 632) as set forth in US Patent No. 8,496,934 or 7,888,486, anti-GDF8 antibodies (*e.g.*, OGD1.0.0, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO: 633 and 634) as set forth in US Patent No. 8,992,913; anti-GDF8 Fab molecules as set forth in European Patent No. 1 773 041 B1, and anti-GDF8 antibodies (*e.g.*, C12, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NOs: 635 and 636, C12-N93H, and/or 510C2 having HCVR/LCVR amino acid sequences of SEQ ID NOs: 637 and 638) as set forth in, *e.g.*, US Patent Nos. 7,635,760 and 8,063,188, anti-GDF8 antibodies (e.g. 41C1E4/landogrozumab/ LY2495655, e.g., having HCVR/LCVR amino acid sequences of SEQ ID NO: 639 and 640) as set forth in US Patent No. 7,632,499. In some embodiments, the anti-GDF8 antibody may have the full length heavy chain and full length light chain amino acid sequences of landogrozumab, e.g., SEQ ID Nos 641 and 642, respectively. In some embodiments, the anti-GDF8

antibody may comprise three heavy chain CDRs (HCDRs) and three light chain CDRs (LCDRs) of landogrozumab, for example, by Chothia definition, according to SEQ ID Nos: 643/644/645/646/647/648, respectively.

[00117] In one embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a heavy chain variable region (HCVR) having an amino acid sequence selected from the group consisting of SEQ ID NO:2, 18, 34, 50, 66, 82, 98, 114, 130, 146, 162, 178, 194, 210, 226, 242, 258, 274, 290, 306, 360, and 376, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00118] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a light chain variable region (LCVR) amino acid sequence selected from the group consisting of SEQ ID NO:10, 26, 42, 58, 74, 90, 106, 122, 138, 154, 170, 186, 202, 218, 234, 250, 266, 282, 298, 314, 322, 368, and 384, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00119] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCVR amino acid sequence and a LCVR amino acid sequence, wherein the HCVR/LCVR sequence pair is selected from the group consisting of SEQ ID NO:2/10, 18/26, 34/42, 50/58, 66/74, 82/90, 98/106, 114/122, 130/138, 146/154, 162/170, 178/186, 194/202, 210/218, 226/234, 242/250, 258/266, 274/282, 290/298, 306/314, 114/322, 360/368, and 376/384.

[00120] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCVR amino acid sequence and a LCVR amino acid sequence, wherein the HCVR/LCVR sequence pair is selected from the group consisting of (HCVR/LCVR): 21-E5 (SEQ ID NO:34/42); 21-B9 (SEQ ID NO:18/26); 21-E9 (SEQ ID NO:98/106); 21-A2 (SEQ ID NO:2/10); 22-D3 (SEQ ID NO:50/58); 22-E6 (SEQ ID NO:66/74); 22-G10 (SEQ ID NO:82/90); 1A2 (SEQ ID NO:226/234); 20B12 (SEQ ID NO:274/282); 58C8 (SEQ ID NO:242/250); 19F2 (SEQ ID NO:258/266); 8D12-1 (SEQ ID NO:114/122); 4E3-7 (SEQ ID NO:194/202); 9B11-12 (SEQ ID NO:162/170); 4B9 (SEQ ID NO:226/234); 1H4-5 (SEQ ID NO:210/218); 9B4-3 (SEQ ID NO:178/186); 3E2-1 (SEQ ID NO:290/298); 4G3-25 (SEQ ID NO:306/314); 4B6-6 (SEQ ID NO:130/138); H4H1657N2 (SEQ ID NO:360/368); H4H1669P (SEQ ID NO:376/384).

[00121] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a heavy chain complementarity determining region 3 (HCDR3) domain

and a light chain CDR3 (LCDR3) domain, wherein the HCDR3 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:8, 24, 40, 56, 72, 88, 104, 120, 136, 152, 168, 184, 200, 216, 232, 248, 264, 280, 296, 312, 366, and 382, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, and the LCDR3 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:16, 32, 48, 64, 80, 96, 112, 128, 144, 160, 176, 192, 208, 224, 240, 256, 272, 288, 304, 320, 328, 374, and 390, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity. In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises an HCDR3/LCDR3 amino acid sequence pair selected from the group consisting of SEQ ID NO:8/16, 24/32, 40/48, 56/64, 72/80, 88/96, 104/112, 120/128, 136/144, 152/160, 168/176, 184/192, 200/208, 216/224, 232/240, 248/256, 264/272, 280/288, 296/304, 312/320, 120/328, 366/374, and 382/390.

[00122] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises heavy chain CDR1 (HCDR1) and CDR2 (HCDR2) domains and light chain CDR1 (LCDR1) and CDR2 (LCDR2) domains, wherein the HCDR1 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:4, 20, 36, 52, 68, 84, 100, 116, 132, 148, 164, 180, 196, 212, 228, 244, 260, 276, 292, 308, 362, and 378, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; the HCDR2 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:6, 22, 38, 54, 70, 86, 102, 118, 134, 150, 166, 182, 198, 214, 230, 246, 262, 278, 294, 310, 364, and 380, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; the LCDR1 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:12, 28, 44, 60, 76, 92, 108, 124, 140, 156, 172, 188, 204, 220, 236, 252, 268, 284, 300, 316, 324, 370, and 386, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity and the LCDR2 domain has an amino acid sequence selected from the group consisting of SEQ ID NO:14, 30, 46, 62, 78, 94, 110, 126, 142, 158, 174, 190, 206, 222, 238, 254, 270, 286, 302, 318, 326, 372, and 388, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity. In another embodiment, the HCDR1, HCDR2 and HCDR3 domains have respective amino acid sequence combinations selected from the group consisting of SEQ ID NO:36/38/40, 116/118/120, 228/230/232, 362/364/366, and 378/380/382; and the LCDR1, LCDR2 and

LCDR3 domains have respective amino acid sequence combinations selected from the group consisting of SEQ ID NO:44/46/48, 124/126/128, 236/238/240, 370/372/374, and 386/388/390.

[00123] In yet another embodiment, the heavy and light chain CDR domains of the anti-GDF8 antibody or antigen-binding fragment thereof (HCDR1/HCDR2/HCDR3/LCDR1/LCDR2/LCDR3) have amino acid sequence combinations selected from the group consisting of SEQ ID NO: 36/38/40/44/46/48 (*e.g.*, 21-E5), 116/118/120/124/126/128 (*e.g.*, 8D12), 228/230/232/236/238/240 (*e.g.*, 1A2), 362/364/366/370/372/374 (*e.g.*, H4H1657N2), and 378/380/382/386/388/390 (*e.g.*, H4H1669P).

[00124] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises heavy and light chain CDR domains contained within heavy and light chain variable region (HCVR/LCVR) amino acid sequence pairs selected from the group consisting of SEQ ID NO: 2/10, 18/26, 34/42, 50/58, 66/74, 82/90, 98/106, 114/122, 130/138, 146/154, 162/170, 178/186, 194/202, 210/218, 226/234, 242/250, 258/266, 274/282, 290/298, 306/314, 114/322, 360/368, and 376/384.

[00125] Methods and techniques for identifying CDRs within HCVR and LCVR amino acid sequences are well known in the art and can be used to identify CDRs within the specified HCVR and/or LCVR amino acid sequences disclosed herein. Exemplary conventions that can be used to identify the boundaries of CDRs include, *e.g.*, the Kabat definition, the Chothia definition, and the AbM definition. In general terms, the Kabat definition is based on sequence variability, the Chothia definition is based on the location of the structural loop regions, and the AbM definition is a compromise between the Kabat and Chothia approaches. See, *e.g.*, Kabat, "Sequences of Proteins of Immunological Interest," National Institutes of Health, Bethesda, Md. (1991); Al-Lazikani et al., J. Mol. Biol. 273:927-948 (1997); and Martin et al., Proc. Natl. Acad. Sci. USA 86:9268-9272 (1989). Public databases are also available for identifying CDR sequences within an antibody.

[00126] In one embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCVR having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:1, 17, 33, 49, 65, 81, 97, 113, 129, 145, 161, 177, 193, 209, 225, 241, 257, 273, 289, 305, 359, and 375, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00127] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a LCVR having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:9, 25, 41, 57, 73, 89, 105, 121, 137, 153, 169, 185, 201, 217, 233, 249, 265, 281, 297, 313, 321, 367, and 383, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00128] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCVR/LCVR pair having amino acid sequences encoded by a nucleic acid molecule pair selected from the group consisting of SEQ ID NO: 1/9, 17/25, 33/41, 49/57, 65/73, 81/89, 97/105, 113/121, 129/137, 145/153, 161/169, 177/185, 193/201, 209/217, 225/233, 241/249, 257/265, 273/281, 289/297, 305/313, 113/321, 359/367, and 375/383.

[00129] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCDR3 domain having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:7, 23, 39, 55, 71, 87, 103, 119, 135, 151, 167, 183, 199, 215, 231, 247, 263, 279, 295, 311, 365, and 381, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, and a LCDR3 domain having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:15, 31, 47, 63, 79, 95, 111, 127, 143, 159, 175, 191, 207, 223, 239, 255, 271, 287, 303, 319, 327, 373, and 389, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity. In one embodiment, the HCDR3/LCDR3 domain pair has amino acid sequences encoded by a nucleic acid sequence pair selected from the group consisting of SEQ ID NO:7/15, 23/31, 39/47, 55/63, 71/79, 87/95, 103/111, 119/127, 135/143, 151/159, 167/175, 183/191, 199/207, 215/223, 231/239, 247/255, 263/271, 279/287, 295/303, 311/319, 119/327, 365/373, and 381/389.

[00130] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises HCDR1 and HCDR2 domains, and LCDR1 and LCDR2 domains, wherein the HCDR1 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:3, 19, 35, 51, 67, 83, 99, 115, 131, 147, 163, 179, 195, 211, 227, 243, 259, 275, 291, 307, 361, and 377, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, the HCDR2 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:5, 21, 37, 53,

69, 85, 101, 117, 133, 149, 165, 181, 197, 213, 229, 245, 261, 277, 293, 309, 363, and 379, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, the LCDR1 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:11, 27, 43, 59, 75, 91, 107, 123, 139, 155, 171, 187, 203, 219, 235, 251, 267, 283, 299, 315, 323, 369, and 385, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, and the LCDR2 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:13, 29, 45, 61, 77, 93, 109, 125, 141, 157, 173, 189, 205, 221, 237, 253, 269, 285, 301, 317, 325, 371, and 387.

[00131] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof comprises heavy and light chain CDR domains (HCDR1/HCDR2/HCDR3/LCDR1/LCDR2/LCDR3) having amino acid sequence combinations encoded by a nucleic acid sequence set of SEQ ID NO:35/37/39/43/45/47, 115/117/119/123/125/127, 227/229/231/235/237/239, 361/363/365/369/371/373, or 377/379/381/385/387/389.

[00132] In a preferred embodiment, the anti-GDF8 antibody or antigen-binding fragment thereof that specifically binds GDF8 comprises the HCDRs of a heavy chain variable region (HCVR) comprising SEQ ID NO:360 and the LCDRs of a light chain variable region (LCVR) comprising SEQ ID NO:368. In another embodiment, the anti-GDF8 antibody or antigen-binding fragment that specifically binds GDF8 comprises three HCDRs comprising SEQ ID NO:362, SEQ ID NO:364, and SEQ ID NO:366 and three LCDRs comprising SEQ ID NO:370, SEQ ID NO:372, and SEQ ID NO:374.

[00133] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof is a fully human or humanized antibody or antibody fragment that binds GDF8 with an affinity (expressed as a dissociation constant, "KD") of about 1 nM or less, as measured by surface plasmon resonance assay (for example, BIACORE™). In certain embodiments, the antibody of the invention exhibits a KD of about 700 pM or less; about 500 pM or less; about 320 pM or less; about 160 pM or less; about 100 pM or less; about 50 pM or less; about 10 pM or less; or about 5 pM or less.

[00134] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof is a fully human or humanized monoclonal antibody (mAb) that specifically binds and inhibits human GDF8 and exhibits an IC50 of less than or equal to about 10 nM; about

5 nM or less; about 3 nM or less; about 2 nM or less; about 1 nM or less; about 500 pM or less; or about 200 pM or less, as measured by GDF8 inducible luciferase assay.

[00135] In one embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof has a modified glycosylation pattern. In some applications, modification to remove undesirable glycosylation sites may be useful, or an antibody lacking a fucose moiety present on the oligosaccharide chain, for example, to increase antibody dependent cellular cytotoxicity (ADCC) function (Shield et al. (2002) JBC 277:26733). In other applications, modification of a galactosylation can be made in order to modify complement dependent cytotoxicity (CDC).

[00136] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof competes for specific binding to GDF8 with another antibody comprising a HCDR1/HCDR2/HCDR3/LCDR1/LCDR2/LCDR3 domain combination having amino acid sequences selected from the group consisting of SEQ ID NO:36/38/40/44/46/48, 116/118/120/124/126/128, 228/230/232/236/238/240, 362/364/366/370/372/374, or 378/380/382/386/388/390. In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof competes for specific binding to GDF8 with another antibody comprising a HCVR/LCVR amino acid sequence pair of SEQ ID NO:2/10, 18/26, 34/42, 50/58, 66/74, 82/90, 98/106, 114/122, 130/138, 146/154, 162/170, 178/186, 194/202, 210/218, 226/234, 242/250, 258/266, 274/282, 290/298, 306/314, 114/322, 360/368, or 376/384.

[00137] In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof recognizes the epitope on GDF8 that is recognized by another antibody comprising a HCDR1/HCDR2/HCDR3/LCDR1/LCDR2/LCDR3 domain combination having amino acid sequences selected from the group consisting of SEQ ID NO: 36/38/40/44/46/48, 116/118/120/124/126/128, 228/230/232/236/238/240, 362/364/366/370/372/374, or 378/380/382/386/388/390. In another embodiment, an anti-GDF8 antibody or antigen-binding fragment thereof recognizes the epitope on GDF8 that is recognized by another antibody comprising a HCVR/LCVR amino acid sequence pair of SEQ ID NO:2/10, 18/26, 34/42, 50/58, 66/74, 82/90, 98/106, 114/122, 130/138, 146/154, 162/170, 178/186, 194/202, 210/218, 226/234, 242/250, 258/266, 274/282, 290/298, 306/314, 114/322, 360/368, or 376/384.

Activin A inhibitors

[00138] The present invention includes methods for altering body composition, inducing a reduction in fat mass, and/or increasing lean mass in a subject, and methods for treating a

disease or disorder characterized by increased fat mass in a subject comprising administering a composition comprising an effective amount of an Activin A inhibitor to the subject.

[00139] As used herein, an "Activin A inhibitor" is any agent that binds to or interacts with human Activin A and interferes with or inhibits the normal biological function of Activin A *in vitro* or *in vivo*. Non-limiting examples of categories of Activin A inhibitors include small molecule Activin A antagonists, nucleic acid-based inhibitors of Activin A expression or activity (*e.g.*, siRNA or antisense), peptide-based molecules that specifically interact with Activin A (*e.g.*, peptibodies), receptor molecules that specifically interact with Activin A, Activin A -binding scaffold molecules, proteins comprising a ligand-binding portion of a receptor that specifically binds Activin A, and anti- Activin A aptamers or portions thereof. In a preferred embodiment, an Activin A inhibitor that can be used in the context of the present invention is an anti-Activin A antibody or antigen-binding fragment thereof that specifically binds human Activin A. Anti-Activin A antibodies include neutralizing and/or blocking antibodies. The inhibition caused by anti-Activin A neutralizing and/or blocking antibodies need not be complete, as long as it is detectable using appropriate assays.

[00140] Activins are homo- and hetero-dimeric molecules comprising βA and/or βB subunits. The βA subunit has the amino acid sequence of SEQ ID NO:617 and the βB subunit has the amino acid sequence of SEQ ID NO: 619. Activin A is a homodimer of two βA subunits; Activin B is a homodimer of two βB subunits; and Activin AB is a heterodimer of one βA subunit and one βB subunit. An anti-Activin A antibody or antigen-binding fragment thereof specifically binds the βA subunit. Since the βA subunit is found in both Activin A and Activin AB molecules, an "anti-Activin A antibody or antigen-binding fragment thereof" can specifically bind Activin A, as well as Activin AB (by virtue of its interaction with the βA subunit). Therefore, an anti-Activin A antibody or antigen-binding fragment thereof specifically binds Activin A, or Activin A and Activin AB, but does not bind other ActRIIB ligands, such as Activin B, GDF3, GDF8, BMP2, BMP4, BMP7, BMP9, BMP10, GDF11, Nodal, *etc.*

[00141] In some embodiments, an anti-Activin A antibody or antigen-binding fragment thereof is employed as set forth in US Patent No. 9,718,881. Exemplary anti-Activin A antibodies that can be used in the compositions and methods of the invention include, *e.g.*, the fully-human anti-Activin antibody H4H10446P2, also known as REGN2477, (*e.g.*, an anti-Activin A antibody comprising the heavy and light chain variable regions having

amino acid sequences SEQ ID NO: 162 and SEQ ID NO: 146, respectively, as set forth in US Patent No. 9,718,881).

[00142] Table 2 sets forth heavy and light chain variable region amino acid sequence pairs of selected anti-Activin A antibodies and their corresponding antibody identifiers that can be used in the compositions and methods of the invention. The corresponding nucleic acid sequence identifiers are set forth in Table 3.

Table 2: anti-Activin A Amino Acid Sequence Identifiers

	SEQ ID NOs:							
Antibody Designation	HCVR	HCDR1	HCDR2	HCDR3	LCVR	LCDR1	LCDR2	LCDR3
H4H10423P	393	395	397	399	401	403	405	407
H4H10424P	409	411	413	415	417	419	421	423
H4H10426P	425	427	429	431	433	435	437	439
H4H10429P	441	443	445	447	449	451	453	455
H4H10430P	457	459	461	463	465	467	469	471
H4H10432P2	473	475	477	479	481	483	485	487
H4H10433P2	489	491	493	495	481	483	485	487
H4H10436P2	497	499	501	503	481	483	485	487
H4H10437P2	505	507	509	511	481	483	485	487
H4H10438P2	513	515	517	519	481	483	485	487
H4H10440P2	521	523	525	527	481	483	485	487
H4H10442P2	529	531	533	535	537	539	541	543
H4H10445P2	545	547	549	551	537	539	541	543
H4H10446P2	553	555	557	559	537	539	541	543
H4H10447P2	561	563	565	567	537	539	541	543
H4H10448P2	569	571	573	575	537	539	541	543
H4H10452P2	577	579	581	583	537	539	541	543
H4H10468P2	585	-587	589	591	537	539	541	543
H2aM10965N	593	595	597	599	601	603	605	607

Table 3: anti-Activin A Nucleic Acid Sequence Identifiers

SEQ ID NOs:								
Antibody Designation	HCVR	HCDR1	HCDR2	HCDR3	LCVR	LCDR1	LCDR2	LCDR3
H4H10423P	392	394	396	398	400	402	404	406
H4H10424P	408	410	412	414	416	418	420	422
H4H10426P	424	426	428	430	432	434	436	438
H4H10429P	440	441	444	446	448	450	452	454
H4H10430P	456	458	460	462	464	466	468	470
H4H10432P2	472	474	476	478	480	482	484	486
H4H10433P2	488	490	492	494	480	482	484	486
H4H10436P2	496	498	500	502	480	482	484	486
H4H10437P2	504	506	508	510	480	482	484	486
H4H10438P2	512	514	516	518	480	482	484	486
H4H10440P2	520	522	524	526	480	482	484	486
H4H10442P2	528	530	532	534	536	538	540	542
H4H10445P2	544	546	548	550	536	538	540	542
H4H10446P2	552	554	556	558	536	538	540	524
H4H10447P2	560	562	564	566	536	538	540	542
H4H10448P2	568	570	572	574	536	538	540	542
H4H10452P2	576	578	580	582	536	538	540	542
H4H10468P2	584	586	588	590	536	538	540	542
H2aM10965N	592	594	596	598	600	602	604	606

[00143] In one embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a heavy chain variable region (HCVR) having an amino acid sequence selected from the group consisting of SEQ ID NO: 393, 409, 425, 441, 457, 473, 489, 497, 505, 513, 521, 529, 545, 553, 561, 569, 577, 585, and 593, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00144] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a light chain variable region (LCVR) having an amino acid sequence selected from the group consisting of SEQ ID NO: 401, 417, 433, 449, 465, 481, 537, and

601, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00145] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a HCVR and LCVR (HCVR/LCVR) amino sequence pair selected from the group consisting of SEQ ID NO: 393/401, 409/417, 425/433, 441/449, 457/465, 473/481, 489/481, 497/481, 505/481, 513/481, 521/481, 529/537, 545/537, 553/537, 561/537, 569/537, 577/537, 585/537, and 593/601.

[00146] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a heavy chain CDR3 (HCDR3) domain having an amino acid sequence selected from the group consisting of SEQ ID NO: 399, 415, 431, 447, 463, 479, 495, 503, 511, 519, 527, 535, 551, 559, 567, 575, 583, 591, and 599, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; and a light chain CDR3 (LCDR3) domain having an amino acid sequence selected from the group consisting of SEQ ID NO: 407, 423, 439, 455, 471, 487, 543, and 607, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00147] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a HCDR3/LCDR3 amino acid sequence pair selected from the group consisting of SEQ ID NO: 399/407, 415/423, 431/439, 447/455, 463/471, 479/487, 495/487, 503/487, 511/487, 519/487, 527/487, 535/543, 551/543, 559/543, 567/543, 575/543, 583/543, 591/543, and 599/607.

[00148] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a heavy chain CDR1 (HCDR1) domain having an amino acid sequence selected from the group consisting of SEQ ID NO: 395, 411, 427, 443, 459, 475, 491, 499, 507, 515, 523, 531, 547, 555, 563, 571, 579, 587, and 595, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; a heavy chain CDR2 (HCDR2) domain having an amino acid sequence selected from the group consisting of SEQ ID NO: 397, 413, 429, 445, 461, 477, 493, 501, 509, 517, 525, 533, 549, 557, 565, 573, 581, 589, and 597, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; a light chain CDR1 (LCDR1) domain having an amino acid sequence selected from the group consisting of SEQ ID NO: 403, 419, 435, 451, 467, 483, 539, and 603, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity; and a light chain CDR2 (LCDR2) domain having an amino

acid sequence selected from the group consisting of SEQ ID NO: 405, 421, 437, 453, 469, 485, 541, and 605, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00149] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises HCDR1-HCDR2-HCDR3-LCDR1-LCDR2-LCDR3 domains, respectively, having the amino acid sequences selected from the group consisting of: SEQ ID NOs: 395-397-8-403-405-407; 411-413-415-419-421-423; 36-429-431-435-437-439; 443-445-447-451-453-455; 459-461-463-467-469-471; 475-477-479-483-485-487; 491-493-495-483-485-487; 499-501-503-483-485-487; 507-509-511-483-485-487; 515-517-519-483-485-487; 523-525-527-483-485-487; 531-533-535-539-541-543; 547-549-551-539-541-543; 555-557-559-539-541-543 (H4H10446P2); 563-565-567-539-541-543; 571-573-575-539-541-543; 579-581-583-539-541-543; 587-589-591-539-541-543; and 595-597-599-603-605-607.

[00150] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises the heavy and light chain CDR domains contained within heavy and light chain variable region (HCVR/LCVR) sequence pairs selected from the group consisting of SEQ ID NO: 393/401, 409/417, 425/433, 441/449, 457/465, 473/481, 489/481, 497/481, 505/481, 513/481, 521/481, 529/537, 545/537, 553/537,561/537, 569/537, 577/537,585/537, and 593/601.

[00151] In one embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a HCVR having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:392, 408, 424, 440, 456, 472, 488, 496, 504, 512, 520, 528, 544, 552, 560, 568, 576, 584, and 592, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00152] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a LCVR having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO: 400, 416, 432, 448, 464, 480, 536, and 600, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00153] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a HCVR and a LCVR, wherein the HCVR/LCVR pair has amino acid sequences encoded by a nucleic acid sequence pair selected from the group consisting of SEQ ID NO: 392/400, 408/416, 424/432, 440/448, 456/464, 472/480, 488/480, 496/480,

504/480, 512/480, 520/480, 528/536, 544/536, 552/536, 560/536, 568/536, 576/536, 584/536, and 592/600.

[00154] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises a HCDR3 domain having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:398, 414, 430, 446, 462, 478, 498, 502, 510, 518, 526, 534, 550, 558, 566, 574, 582, 590, and 598, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, and a LCDR3 domain having an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:406, 422, 435, 454, 470, 486, 542, and 606, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity. In one embodiment, the HCDR3/LCDR3 domain set has amino acid sequences encoded by a nucleic acid sequence pair selected from the group consisting of SEQ ID NO:398/406, 414/422, 430/438, 446/454, 462/470, 478/486, 494/486, 502/486, 510/486, 518/486, 526/486, 534/542, 550/542, 558/542, 566/542, 574/542, 582/542, 590/542, and 598/606.

[00155] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises HCDR1 and HCDR2 domains, and LCDR1 and LCDR2 domains, wherein the HCDR1 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:394, 410, 426, 442, 458, 474, 490, 498, 506, 514, 522, 530, 546, 554, 562, 570, 578, 586, and 594, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, the HCDR2 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:396, 412, 428, 444, 460, 476, 492, 500, 508, 516, 524, 532, 548, 556, 564, 572, 580, 588, and 596, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, the LCDR1 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:402, 418, 434, 450, 466, 482, 538, and 602, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity, and the LCDR2 domain has an amino acid sequence encoded by a nucleic acid sequence selected from the group consisting of SEQ ID NO:404, 420, 436, 452, 468, 484, 540, and 604, or a substantially similar sequence thereof having at least 90%, at least 95%, at least 98% or at least 99% sequence identity.

[00156] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof comprises heavy and light chain CDR (HCDR1, HCDR2, HCDR3, LCDR1, LCDR2, LCDR3) domains having amino acid sequences encoded by a nucleic acid sequence set selected from the group consisting of SEQ ID NO:394/396/398/402/404/406, 410/412/414/418/420/422, 426/428/430/434/436/438, 442/444/446/450/452/454, 458/460/462/466/468/470, 474/476/478/482/484/486, 490/492/494/482/484/486, 498/500/502/482/484/486, 506/508/510/482/484/486, 514/516/518/482/484/486, 522/524/526/482/484/486, 530/532/534/538/540/542, 546/548/550/538/540/542, 554/556/558/538/540/542, 562/564/566/538/540/542, 570/572/574/538/540/542, 578/580/582/538/540/542, 586/588/590/538/540/542, and 594/596/598/602/604/606.

[00157] In one embodiment, the anti-Activin A antibody or antigen-binding fragment thereof comprises the HCVR and LCVR (HCVR/LCVR) amino sequence pair of SEQ ID NO: 553/537 and the anti-GDF8 antibody or antigen-binding fragment thereof comprises a HCVR and LCVR (HCVR/LCVR) amino sequence pair of SEQ ID NO: 360/368.

[00158] In another embodiment, the anti-Activin A antibody or antigen-binding fragment thereof comprises HCDR1-HCDR2-HCDR3-LCDR1-LCDR2-LCDR3 domains, respectively, having the amino acid sequences of: SEQ ID NOs: 555-557-559-539-541-543 (H4H10446P2), and the anti-GDF8 antibody or antigen-binding fragment thereof comprises HCDR1-HCDR2-HCDR3-LCDR1-LCDR2-LCDR3 domains, respectively, having the amino acid sequences of: SEQ ID NOs: 362/364/366/370/372/374 (*e.g.*, H4H1657N2).

[00159] In another embodiment, an anti-Activin A antibody or antigen-binding fragment thereof has a modified glycosylation pattern. In some applications, modification to remove undesirable glycosylation sites may be useful, or an antibody lacking a fucose moiety present on the oligosaccharide chain, for example, to increase antibody dependent cellular cytotoxicity (ADCC) function (Shield et al. (2002) JBC 277:26733). In other applications, modification of a galactosylation can be made in order to modify complement dependent cytotoxicity (CDC).

[00160] The fully-human anti-Activin A and/or anti-GDF8 antibodies described herein 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. 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

compositions and methods of the invention use, in additional embodiments, antibodies and antigen-binding fragments thereof that 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 back-mutated to the corresponding germline residue(s) or to a conservative amino acid substitution (natural or non-natural) of the corresponding germline residue(s) (such sequence changes are referred to herein as "germline back-mutations"). A person of ordinary skill in the art, starting with the heavy and light chain variable region sequences described herein, can easily produce numerous antibodies and antigen-binding fragments that comprise one or more individual germline back-mutations or combinations thereof. In certain embodiments, all of the framework and/or CDR residues within the VH and/or VL domains are mutated back to the germline sequence. In other embodiments, only certain residues are mutated back to the 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. Furthermore, the antibodies and antigen-binding fragments used in the compositions and methods of the invention may contain any combination of two or more germline back-mutations within the framework and/or CDR regions, *i.e.*, wherein certain individual residues are mutated back to the germline sequence while certain other residues that differ from the germline sequence are maintained. Once obtained, antibodies and antigen-binding fragments that contain one or more germline back-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. Antibodies and antigen-binding fragments obtained in this general manner are encompassed within the invention.

[00161] The compositions and methods of the invention use, in additional embodiments, anti-GDF8 antibodies and/or anti-Activin A antibodies (or antigen-binding fragments thereof) comprising variants of any of the HCVR, LCVR, and/or CDR amino acid sequences described herein having one or more conservative substitutions. For example, anti-GDF8 antibodies and/or anti-Activin A antibodies used in the compositions and methods of the invention have, in some embodiments, 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 described herein.

Bispecific antibodies

[00162] Bispecific antibodies (bsAbs) combine specificities of two antibodies and simultaneously bind different antigens or epitopes. Two or more antigen-recognizing elements are engineered into a single antibody. In one embodiment of the methods of the invention, the composition comprises an antibody comprising a GDF8-specific binding domain and an Activin A-specific binding domain. The term (antigen)"-specific binding domain," as used herein, includes polypeptides comprising or consisting of: (i) an antigen-binding fragment of an antibody molecule, (ii) a peptide that specifically interacts with a particular antigen (*e.g.*, a peptibody), and/or (iii) a ligand-binding portion of a receptor that specifically binds a particular antigen. For example, included are bispecific antibodies with one arm comprising a first heavy chain variable region/light chain variable region (HCVR/LCVR) pair that specifically binds GDF8 and another arm comprising a second HCVR/LCVR pair that specifically binds Activin A.

[00163] Bispecific antibodies can be prepared according to known methods, including chemical cross-linking, hybrid hybridomas/quadromas, knobs into holes, CrossMab, dual-variable-domain immunoglobulin, recombinant engineering (tandem single chain variable fragments/diabodies), and dock and lock. Other exemplary bispecific formats that can be used in the context of the present invention include, without limitation, *e.g.*, IgG-scFv fusions, dual variable domain (DVD)-Ig, common light chain, CrossFab, (SEED)body, leucine zipper, Duobody, IgG1/IgG2, dual acting Fab (DAF)-IgG, and Mab2 bispecific formats (see, *e.g.*, Klein et al., mAbs 4:6, 1-11 (2012), and references cited therein, for a review of the foregoing formats). Bispecific antibodies can also be constructed using peptide/nucleic acid conjugation, *e.g.*, wherein unnatural amino acids with orthogonal chemical reactivity are used to generate site-specific antibody-oligonucleotide conjugates, which then self-assemble into multimeric complexes with defined composition, valency and geometry. (See, *e.g.*, Kazane et al., J Am Chem Soc. 135(1):340–346 (2013)).

Specific Binding

[00164] The term "specifically binds" or the like, as used herein, means that an antigen-specific binding protein, or an antigen-specific binding domain, forms a complex with a particular antigen characterized by a dissociation constant (K_D) of 500 pM or less, and does not bind other unrelated antigens under ordinary test conditions. "Unrelated antigens" are proteins, peptides or polypeptides that have less than 95% amino acid identity to one another. Methods for determining whether two molecules specifically bind one another are well known in the art and include, for example, equilibrium dialysis,

surface plasmon resonance, and the like. For example, an antigen-specific binding protein or an antigen-specific binding domain, as used in the context of the present invention, includes antibodies or antigen-binding fragments thereof that bind a particular antigen (*e.g.*, GDF8, Activin A) or a portion thereof with a K_D of less than about 500 pM, less than about 400 pM, less than about 300 pM, less than about 200 pM, less than about 100 pM, less than about 90 pM, less than about 80 pM, less than about 70 pM, less than about 60 pM, less than about 50 pM, less than about 40 pM, less than about 30 pM, less than about 20 pM, less than about 10 pM, less than about 5 pM, less than about 4 pM, less than about 2 pM, less than about 1 pM, less than about 0.5 pM, less than about 0.2 pM, less than about 0.1 pM, or less than about 0.05 pM, as measured in a surface plasmon resonance assay.

[00165] Antibody binding (of antigen) can be quantitated in terms of K_D, a measurement of affinity. The lower the K_D value, the higher the binding affinity of the antibody. The term "K_D", as used herein, is intended to refer to the equilibrium dissociation constant of a particular antibody-antigen interaction. Surface plasmon resonance can be used to measure ligand binding, for example, antibody-antigen interaction.

[00166] The term "surface plasmon resonance", as used herein, refers to an optical phenomenon that allows for the analysis of real-time biospecific interactions by detection of alterations in protein concentrations within a biosensor matrix, for example using the BIACORE™ system (Pharmacia Biosensor AB, Uppsala, Sweden and Piscataway, N.J.).

[00167] The term “epitope” includes any determinant, preferably a polypeptide determinant, capable of specific binding to an immunoglobulin or T-cell receptor. In certain embodiments, epitope determinants include chemically active surface groupings of molecules such as amino acids, sugar side chains, phosphoryl groups, or sulfonyl groups, and, in certain embodiments, may have specific three-dimensional structural characteristics, and/or specific charge characteristics. An epitope is a region of an antigen that is bound by an antibody. In certain embodiments, an antibody is said to specifically bind an antigen when it preferentially recognizes its target antigen in a complex mixture of proteins and/or macromolecules. For example, an antibody is said to specifically bind an antigen when the K_D is less than or equal to 10^{-8} M, less than or equal to 10^{-9} M, or less than or equal to 10^{-10} M.

Preparation of Human Antibodies

[00168] Methods for generating monoclonal antibodies, including fully human monoclonal antibodies are known in the art. Any such known methods can be used in the context of the invention to make human antibodies that specifically bind to GDF8 and/or to Activin A.

[00169] Using VELOCIMMUNE™ technology or any other known method for generating monoclonal antibodies, high affinity chimeric antibodies to GDF8 and/or Activin A are initially isolated having a human variable region and a mouse constant region. As in the experimental section below, the antibodies are characterized and selected for desirable characteristics, including affinity, selectivity, epitope, etc. 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.

[00170] In general, the antibodies used in the methods of the instant invention possess very high affinities, typically possessing K_D of from about 10^{-12} through about 10^{-9} M, when measured by binding to antigen either immobilized on solid phase or in solution phase.

Pharmaceutical Compositions and Methods of Administration

[00171] The present invention includes methods for altering body composition of a subject. As used herein, the phrase "altering body composition" refers to a change in one or more of lean mass, fat mass, and/or bone mass in a subject. In some embodiments, body composition in a subject may be altered by administering an effective amount of a GDF8 inhibitor and an Activin A inhibitor to the subject. Lean mass may be, for example, thigh muscle volume, appendicular lean body mass, or total lean mass, etc. In some aspects, the thigh muscle volume may refer to thigh muscle tissue volume excluding intramuscular adipose tissue and large vessels. In some aspects, the thigh muscle volume may refer to thigh muscle tissue volume including intramuscular adipose tissue and large vessels. In some aspects, the appendicular lean body mass may be calculated by, for example, aLBM equation. In some aspects, the appendicular lean mass may be calculated by the sum of lean mass of arms and legs. Fat mass may be, for example, total fat mass, android fat mass, sum of intramuscular and perimuscular adipose tissue (IMAT), subcutaneous adipose tissue volume, sum of fat mass of arms and legs, thigh intramuscular adipose tissue, etc. Bone mass may be, for example, total bone mineral density(BMD)

mass, total bone mineral content (BMC) mass, etc. In some embodiments, alteration of body composition comprises an increase in muscle mass and/or a reduction of fat mass. In some embodiments, alteration of body composition comprises an increase in muscle mass and a reduction of fat mass simultaneously. In some embodiments, alteration of body composition comprises an increase in muscle mass and a reduction of fat mass simultaneously, without reduction in bone mass. In some embodiments, alteration of body composition comprises an increase in bone mineral content mass. In some embodiments, alteration of body composition comprises a decrease in total fat mass, android fat mass, and/or subcutaneous fat mass. In some embodiments, alteration of body composition comprises a decrease in total fat mass, android fat mass, and/or subcutaneous fat mass, without a reduction in thigh intramuscular adipose tissue volume.

[00172] The present invention includes methods for altering body composition, for example, inducing a reduction in fat mass in a subject and methods for treating a disease or disorder characterized by increased fat mass, comprising administering a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor to the subject. The first and second compositions can be administered concurrently or sequentially to the subject. The first and second compositions can also be combined into a third composition prior to administration. Thus, in certain embodiments, a composition comprising both a GDF8 inhibitor and an Activin A inhibitor can be administered to a subject. The GDF8 inhibitor in such a composition can, for example, be an anti-GDF8 antibody. The Activin A inhibitor in such a composition can, for example, be an anti-Activin A antibody.

[00173] 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. Any of the foregoing mixtures may be appropriate in treatments and therapies in accordance with the invention, provided that the active ingredient in the formulation is not inactivated by the formulation and the formulation is physiologically compatible and tolerable with the route

of administration. See also Powell et al. "Compendium of excipients for parenteral formulations" PDA (1998) J Pharm Sci Technol 52:238-311.

[00174] Various delivery systems are known and can be used to administer the pharmaceutical compositions of the present 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 compositions 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.

[00175] 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.

[00176] 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.

[00177] In certain situations, the pharmaceutical compositions of the present invention 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.

[00178] 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.

[00179] 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

[00180] The amount of active ingredient (*e.g.*, anti-GDF8 antibody and/or anti-Activin A antibody) that can be administered to a subject is, generally, a therapeutically effective amount. The term "effective amount" is a concentration or amount of an active ingredient, for example, an antibody or antigen-binding fragment of an antibody, which results in achieving a particular stated purpose. The term "effective amount" is used interchangeably with the term "therapeutically effective amount" and signifies a concentration or amount of an active ingredient, for example, an antibody or antigen-binding fragment thereof, which is effective for achieving a stated therapeutic effect. The (therapeutically) effective amount may be determined empirically.

[00181] As used herein, the phrase "therapeutically effective amount" or "effective amount" means a dose of antigen-specific binding proteins and/or antigen-binding molecules (*e.g.*, antibodies) that results in a detectable decrease in fat mass. The effective amount may also, in certain embodiments, result in an increase in one or more of the following parameters: body weight, muscle mass (*e.g.*, tibialis anterior [TA] muscle mass, gastrocnemius [GA] muscle mass, quadriceps [Quad] muscle mass, appendicular lean body mass, etc.), muscle volume (*e.g.*, thigh muscle volume), muscle strength/power, and/or muscle function, and glucose tolerance.

[00182] A "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a decrease in total fat mass of at least about 2% to 8%, at least 2.5% to 6%, at least 3% to 4%, or at least about 2.0%, at least about 2.5%, at least about 3.0%, or at least about 3.5%, or more. For example, a "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a decrease in total fat mass of at least about 3.5% or more.

[00183] In some embodiments, a "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a decrease in android fat mass of at least about 2% to 8%, at least 2.5% to 6%, at least 3% to 4%, or at least about 2.0%, at least about 2.5%, at least about 3.0%, or at least about 3.5%, or more. For example, a

"therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a decrease in android fat mass of at least about 3.5%.

[00184] In certain embodiments, the amount also results in an increase in TA or GA muscle mass of at least 2%, 5%, 10%, 15%, 20%, 25%, 30%, 40%, 50%, 60% or more, compared to control treated subjects.

[00185] In some embodiments, a "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a increase in thigh muscle volume of at least about 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more

[00186] In some embodiments, a "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a increase in total lean body mass of at least about 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more

[00187] In some embodiments, a "therapeutically effective amount" or "effective amount" of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or an Activin A inhibitor (*e.g.*, anti-Activin A antibody) includes, *e.g.*, an amount of GDF8 inhibitor and/or Activin A inhibitor that, when administered to a subject, causes a increase in appendicular lean body mass of at least about 2% to 8%, 2.5% to 6%, 3% to 4%, or at least 2.0%, at least 2.5%, at least 3.0%, or at least 3.5%, or more.

[00188] In certain embodiments, the amount also results in an increase in TA or GA muscle mass of at least 2%, 3%, 5%, 10%, 15%, 20%, 25% or more, compared to control treated subjects.

[00189] In certain embodiments, a (therapeutically) effective amount of an anti-GDF8 antibody, anti-Activin A antibody, or bispecific antibody that specifically binds GDF8 and Activin A 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 150 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 respective antibody. The dose may vary depending upon the age and the size of a subject to be administered, target disease, conditions, route of administration, and the like. Depending on the severity of the condition, the frequency and the duration of the treatment can be adjusted.

[00190] The amount of antibody (*e.g.*, anti-GDF8 antibody, anti-Activin A antibody, or bispecific antibody that specifically binds GDF8 and Activin A) 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-GDF8 antibody, anti-Activin A antibody, and/or anti-GDF8/anti-Activin A bispecific antibody in the first, second, or third composition administered per the methods of the invention may be administered to a patient at a dose of about 0.0001 to about 50 mg/kg of patient body weight (*e.g.* 0.5 mg/kg, 1.0 mg/kg, 1.5 mg/kg, 2.0 mg/kg, 2.5 mg/kg, 3.0 mg/kg, 3.5 mg/kg, 4.0 mg/kg, 4.5 mg/kg, 5.0 mg/kg, 5.5 mg/kg, 6.0 mg/kg, 6.5 mg/kg, 7.0 mg/kg, 7.5 mg/kg, 8.0 mg/kg, 8.5 mg/kg, 9.0 mg/kg, 9.5 mg/kg, 10.0 mg/kg, 10.5 mg/kg, 11.0 mg/kg, 11.5 mg/kg, etc.).

[00191] The effective amount of a GDF8 inhibitor (*e.g.*, an anti-GDF8 antibody) may, in certain embodiments, comprises a dosing regimen selected from the group consisting of at least 0.1 mg/kg to about 10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100 mg/kg. The effective amount of an Activin A inhibitor (*e.g.*, an anti-Activin A antibody) may, in certain embodiments, comprise a dosing regimen selected from the group consisting of at least 0.1 mg/kg to about 10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100mg/kg.

[00192] The effective amount of a GDF8 inhibitor (*e.g.*, an anti-GDF8 antibody) may, in additional embodiments, comprise a dosing regimen selected from a group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1 to about 5 mg/kg body weight. The effective amount of an Activin A inhibitor (*e.g.*, an anti-Activin A antibody) may, in additional embodiments, comprise a dosing regimen selected from a group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1

to about 5 mg/kg body weight. In one specific aspect, the effective amount of an anti-GDF8 antibody is from about 2 mg/kg-10 mg/kg, 4 mg/kg-8 mg/kg, or about 6 mg/kg body weight of subject, and the effective amount of the anti-Activin A antibody is from 0.5 mg/kg-15 mg/kg, 2 mg/kg- 12 mg/kg, about 3 mg/kg, or about 10 mg/kg body weight of a subject.

[00193] The first, second, and third compositions administered per methods of the invention may, in certain embodiments, comprise equal amounts of GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) and/or Activin A inhibitor (*e.g.*, anti-Activin A antibody). Alternatively, the amount of GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) in the composition may be less than or greater than the amount of Activin A inhibitor (*e.g.*, an anti-Activin A antibody). The effective amount of a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody) may be lower, when in combination with an Activin A inhibitor (*e.g.*, an anti-Activin A antibody), than in a separate composition. The effective amount of an Activin A inhibitor (*e.g.*, an anti-Activin A antibody) may be lower, when in combination with a GDF8 inhibitor (*e.g.*, anti-GDF8 antibody), than in a separate composition. A person of ordinary skill in the art, using routine experimentation, will be able to determine the appropriate amounts of the individual components in the compositions necessary to produce a desired therapeutic effect.

Aspects of the Disclosure

[00194] The disclosure provides compositions, kits, and methods of using GDF8 inhibitors and Activin A inhibitors to reduce fat mass (induce a reduction in fat mass) in a subject. The disclosure also provides compositions, kits, and methods of using GDF8 inhibitors and Activin A inhibitors to treat diseases, disorders, and/or conditions associated with or characterized by increased fat mass in a subject. In preferred embodiments, the GDF8 inhibitor is an antibody or antigen-binding fragment thereof that specifically binds GDF8.

Therapeutic Methods

[00195] The present invention includes methods for altering body composition, for example, methods for inducing a reduction in fat mass in a subject, methods for increasing muscle mass in a subject, and methods for treating a disease or disorder characterized by increased fat mass, by specifically binding GDF8 and/or Activin A. For example, the present invention includes methods for inducing a reduction in fat mass in a subject, inducing an increase in muscle mass in a subject, and methods for treating a disease or disorder characterized by increased fat mass in a subject, by administering to the subject i)

a composition comprising an anti-GDF8 antibody and a composition comprising an anti-Activin A antibody or ii) a composition comprising both an anti-GDF8 antibody and an anti-Activin A antibody or iii) a composition comprising a bispecific antibody comprising a first variable domain comprising a HCVR/LCVR pair that specifically binds GDF8 and a second variable domain comprising a HCVR/LCVR pair that specifically binds Activin A. Any of the GDF8 inhibitors and/or Activin A inhibitors disclosed or referred to herein can be used in the context of these aspects of the invention.

[00196] In methods comprising administering a GDF8 inhibitor and an Activin A inhibitor to a subject, the GDF8 inhibitor (for example, an anti-GDF8 antibody) and the Activin A inhibitor (for example, an anti-Activin A antibody) may be administered to the subject at the same or substantially the same time, *e.g.*, in a single therapeutic dosage (third composition) or in two separate dosages (first and second compositions), which are administered simultaneously or within less than about 5 minutes of one another. Alternatively, the GDF8 inhibitor and the Activin A inhibitor (first and second compositions) may be administered to the subject sequentially, *e.g.*, in separate therapeutic dosages separated in time from one another by more than about 5 minutes.

[00197] The reduction of fat mass in the subject of the methods according to the invention can be a reduction in total fat mass as measured by DXA (Dual-energy X-ray absorptiometry).

[00198] In another embodiment, the reduction of fat mass in the subject of the methods according to the invention is a reduction in android fat mass (*i.e.*, visceral fat associated with the upper/central body) as measured by DXA (Dual-energy X-ray absorptiometry). In android obesity, the subject stores fat around his or her abdominal region. Android obesity can also be manifested in other areas of the upper trunk like the upper chest (front or back) nape area of the neck, and even the shoulders. Subjects who are android obese are at greater risk for obesity-related diseases/disorders like heart disease, and metabolic syndrome. The likelihood of developing gout, arterial-related diseases (due to high blood pressure) and many kinds of cancers are also linked to the central type of fat distribution in subjects who exhibit android obesity.

[00199] Body fat assessments are varied in precision and accuracy. Common anthropometric measures include: weight, waist circumference, and skinfold measurements using skin calipers. More complex methods include: bioelectrical impedance analysis (BIA), the BOD POD, and dual-energy X-ray absorptiometry (DEXA or DXA). DXA is especially accurate and valid, because it considers bone mineral content

when estimating body fat and muscle. DEXA scans can evaluate different areas of fat distribution to determine the android/gynoid fat ratio, which is distinct from body mass index. DXA can measure total fat mass, total body muscle mass, visceral fat (fat around the organs) levels, intramuscular fat (fat between the muscles), total bone mineral density, and can even provide regional breakdowns. Finally, DXA can accurately assess the distribution of body fat associated with increased insulin resistance.

Avoidance of Side Effects

[00200] The present invention includes methods for altering body composition, for example, inducing a reduction in fat mass in a subject, and methods for treating a disease or disorder characterized by increased fat mass, comprising administering a GDF8 inhibitor and an Activin A inhibitor to the subject, without causing adverse side effects associated with the administration of molecules which bind multiple (*e.g.*, 3 or more) ActRIIB ligands, for example, as set forth in US Patent No. 8,871,209. For example, the clinical molecule referred to as ACE-031 (Acceleron Pharma, Inc., Cambridge, MA) is a multimer consisting of the extracellular portion of ActRIIB fused to an IgG Fc domain (this molecule is also referred to herein as "ActRIIB-Fc"). ActRIIB-Fc binds GDF8 as well as other ActRIIB ligands such as, *e.g.*, Activin A, Activin B, GDF11, BMP9, BMP10, and TGFβ, and is known to cause various adverse side effects when administered to human patients. For example, administration of ACE-031 to postmenopausal women in a Phase Ib ascending dose study was shown to cause undesired increases in hemoglobin and decreases in FSH levels. In addition, a Phase II study of ACE-031 in pediatric patients with muscular dystrophy was discontinued due to adverse effects including nose and gum bleeding. Dilated blood vessels are also observed in patients treated with ActRIIB-Fc. Effect of ACE-031 in boys with Duchenne muscular dystrophy (DMD) demonstrated trends for increased lean body mass and reduced fat mass but non-muscle-related adverse events contributed to a decision to discontinue the study. (Campbell, *et al.* 2017 Muscle Nerve 55: 458-464). Specifically inhibiting GDF8 and Activin A (*e.g.*, by administering an anti-GDF8 antibody and an anti-Activin A antibody), while not inhibiting other ActRIIB ligands such as Activin B, GDF11, BMP9, BMP10, and TGFβ, results in an increase in a reduction in fat mass, without causing the adverse side effects associated with non-specific Activin-binding agents such as ActRIIB-Fc.

Administration Regimens

[00201] According to certain embodiments of the present invention, multiple doses of the compositions of the present invention (*e.g.*, compositions comprising a GDF8 inhibitor

and/or an Activin A inhibitor, for example, an anti-GDF8 antibody and/or an anti-Activin A antibody, or a bispecific antibody against GDF8 and Activin A), may be administered to a subject over a defined time course. The methods according to this aspect of the invention comprise sequentially administering to a subject multiple doses of the composition(s) of the present invention. As used herein, "sequentially administering" means that each dose of the compositions of the present invention are 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 that comprise sequentially administering to the patient an initial dose of a first and/or a second composition; or a third composition; followed by one or more secondary doses of the first and/or second composition; or the third composition; and optionally followed by one or more tertiary doses of the first and/or second composition; or the third composition.

[00202] The terms "initial dose," "secondary doses," and "tertiary doses," refer to the temporal sequence of administration of the compositions of the present invention. Thus, the "initial dose" is the dose that is administered at the beginning of the treatment regimen (also referred to as the "baseline dose"); the "secondary doses" are the doses that are administered after the initial dose; and the "tertiary doses" are the doses that are administered after the secondary doses. The initial, secondary, and tertiary doses may all contain the same amount of active ingredient(s), *e.g.*, anti-GDF8 antibody and/or anti-Activin A antibody, but will generally differ from one another in terms of frequency of administration. In certain embodiments, however, the amount of active ingredient(s) contained in the initial, secondary and/or tertiary doses will vary from one another (*e.g.*, adjusted up or down as appropriate) during the course of treatment.

[00203] In one exemplary embodiment of the present invention, each secondary and/or tertiary dose is administered 1 to 30 (*e.g.*, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, or more) days after the immediately preceding dose. The phrase "the immediately preceding dose," as used herein, means, in a sequence of multiple administrations, the dose(s) of the compositions of the present invention that are administered to a subject prior to the administration of the very next dose in the sequence, with no intervening doses.

[00204] The methods according to this aspect of the invention may comprise administering to a patient any number of secondary and/or tertiary doses of the compositions of the present invention. 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.

[00205] 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 29 days 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 60 days 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.

[00206] In one embodiment, a subject may be subjected to preliminary DXA, then receive a composition comprising an anti-GDF8 antibody and an anti-Activin A antibody (or a composition comprising an anti-GDF8 antibody and a composition comprising an anti-Activin A antibody), then be subjected to follow-up DXA. If the fat mass is not measurably reduced in the follow-up DXA (in comparison with the preliminary DXA), the subject may receive the composition(s) again. Subsequent dosage amount and frequency of administration can, in an additional embodiment, be varied based on the results of the follow-up DXA.

Combination Therapies

[00207] The methods of the present invention, according to certain embodiments, comprise administering to the subject one or more additional therapeutic agents that may be advantageously combined with the composition comprising a GDF8 inhibitor and/or an Activin A inhibitor. As used herein, the expression "in combination with" means that the additional therapeutic agent(s) is/are administered before, after, or concurrently with a pharmaceutical composition comprising a GDF8 inhibitor and/or an Activin A inhibitor. The term “in combination with” also includes sequential or concomitant administration of a GDF8 inhibitor, an Activin A inhibitor, or both and a second therapeutic agent. The term “therapeutic agent” is also meant to include a specific therapy.

[00208] The additional therapeutic agent may be, *e.g.*, another GDF8 antagonist/inhibitor, another Activin A antagonist/inhibitor, growth factor inhibitors, immunosuppressants,

metabolic inhibitors, enzyme inhibitors, and cytotoxic/cytostatic agents, an IL-1 antagonist (including, *e.g.*, an IL-1 antagonist as set forth in US 6,927,044), an IL-6 antagonist, an IL-6R antagonist (including, *e.g.*, an anti-IL-6R antibody as set forth in US 7,582,298), an IL-13 antagonist, a tumor necrosis factor (TNF) antagonist, an IL-8 antagonist, an IL-9 antagonist, an IL-17 antagonist, an IL-5 antagonist, an IgE antagonist, a CD48 antagonist, an IL-31 antagonist (including, *e.g.*, as set forth in US 7,531,637), a thymic stromal lymphopoietin (TSLP) antagonist (including, *e.g.*, as set forth in US 2011/027468), interferon-gamma (IFNγ) antibiotics, topical corticosteroids, tacrolimus, pimecrolimus, cyclosporine, azathioprine, methotrexate, cromolyn sodium, proteinase inhibitors, systemic corticosteroids, systemic immunotherapy, anti-histamines, chemotherapy, light therapy, or combinations thereof.

[00209] In further embodiments, the invention features a composition, the additional therapeutic agent is selected from the group consisting of (1) 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase inhibitors, such as cerivastatin, atorvastatin, simvastatin, pitavastatin, rosuvastatin, fluvastatin, lovastatin, pravastatin, and the like; (2) inhibitors of cholesterol uptake and/or bile acid re-absorption; (3) niacin, which increases lipoprotein catabolism; (4) fibrates or amphipathic carboxylic acids, which reduce low-density lipoprotein (LDL) level, improve high-density lipoprotein (HDL) and TG levels, and reduce the number of non-fatal heart attacks; and (5) activators of the LXR transcription factor that plays a role in cholesterol elimination such as 22-hydroxycholesterol, or fixed combinations such as ezetimibe plus simvastatin; a statin with a bile resin (*e.g.*, cholestyramine, colestipol, colesevelam), a fixed combination of niacin plus a statin (*e.g.*, niacin with lovastatin); or with other lipid lowering agents such as omega-3-fatty acid ethyl esters (for example, omacor).

[00210] In still further embodiments, the second therapeutic agent is selected from one or more other inhibitors/antagonists of glucagon or an inhibitor/antagonist of the glucagon receptor, as well as inhibitors of other molecules, such as inhibitors of ANGPTL8 (for example, an anti-ANGPTL8 antibody), as well as inhibitors of other molecules, such as ANGPTL3 (for example, an anti-ANGPTL3 antibody), ANGPTL4, ANGPTL5, ANGPTL6, apolipoprotein C-III (also referred to as APOC3; see for example, inhibitors of APOC3 described in US8530439, US7750141, US7598227 and volanesorsen, also referred to as ISIS-APOCIIIRx) and proprotein convertase subtilisin/kexin type 9 (PCSK9), which are involved in lipid metabolism, in particular, cholesterol and/or

triglyceride homeostasis. Inhibitors of these molecules include small molecules, antisense molecules and antibodies that specifically bind to these molecules and block their activity.

[00211] The additional therapeutic agent may, in further embodiments, be selected from the group consisting of analgesics, anti-inflammatory agents, including non-steroidal anti-inflammatory drugs (NSAIDS), such as Cox-2 inhibitors, and the like, so as to ameliorate and/or reduce the symptoms accompanying the underlying condition, if needed.

[00212] The additional therapeutic agent(s) may be administered prior to, concurrent with, or after the administration of the first and/or second; or third composition(s) described herein. For purposes of the present disclosure, such administration regimens may be considered the administration of an anti-GDF8 antibody and/or an anti-Activin A antibody "in combination with" a second therapeutically active component.

EXAMPLES

[00213] 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. Anti-GDF8 alone increased total lean mass up to 3% in clinical studies: sarcopenia phase 2 data

[00214] A randomized, double-blind, placebo-controlled interventional study phase 2 clinical trial was performed in patients 70 years and older including men and postmenopausal women having sarcopenia. Patients were treated 12 weeks with subcutaneous anti-GDF8 antibody REGN1033 (H4H1657N2) alone, at either 100 mg anti-GDF8 antibody, Q4W s.c. (n = 62); 300 mg anti-GDF8 antibody Q4W s.c.(n = 64), or 300 mg anti-GDF8 antibody Q2W s.c. (n =59), or placebo (n = 65). As shown in Figure 1 significant increase in total lean body mass compared to placebo was exhibited in patients after 12 weeks when using REGN1033 alone at each of the three doses, as shown in Figure 1 and Table 4.

[00215] Table 4. Total Lean Mass % Change to Week 12 using anti-GDF8 REGN1033 alone

	Placebo	Anti-GDF8		
		100 Q4W	300 Q4W	300 Q2W
N	65	62	64	59
Baseline Mean	43.6 kg	42.9 kg	42.7	42.8
Difference vs. placebo	–	1.66%	1.78%	2.29%
P value	–	0.0077	0.0043	0.0004

[00216] The effects on strength and function were varied. Anti-GDF8 was generally safe and well-tolerated (reactions, if any, were mild). Table 4 shows patients receiving REGN1033 in either 100 mg or 300 mg doses exhibited significantly increased Total lean mass as % change from placebo to week 12 data. Anti-GDF8 antibody REGN1033 alone increased total lean body mass up to 3% in the study. The 300 mg regimens also resulted in decreases in total and android fat mass.

Example 2. Combination of anti-GDF8 and anti-Activin A first in-human single ascending dose study

[00217] A randomized, double-blind placebo-controlled, ascending dose study was initiated to assess the tolerability and effects on body composition of a combination of an intravenous anti-GDF8 antibody and an anti-Activin A antibody vs. the individual components.

[00218] The primary objective of the study was to assess the safety and tolerability of an anti-Activin A antibody (*e.g.*, H4H10446P2 = REGN2477) alone and combined with an anti-GDF8 antibody (*e.g.*, H4H1657N2 = REGN1033) in healthy postmenopausal women aged 45 to 70 years of age.

[00219] Secondary objectives of the study included: an assessment of the effect of REGN2477 alone, REGN1033 alone, and REGN2477+ REGN1033 in combination on thigh muscle volume as measured by Magnetic Resonance Imaging (MRI), and an assessment of the effects of REGN2477 alone, REGN1033 alone and REGN2477+REGN1033 in combination on total and regional body composition as measured by dual-energy X-ray absorptiometry (DXA)

Study Design

[00220] This study was a randomized, double-blind, placebo-controlled, ascending dose study to assess the safety, tolerability, and pharmacodynamics of intravenous REGN2477 (anti-Activin A) alone and in combination with REGN1033 (anti-GDF8) in healthy postmenopausal women.

[00221] A total of 48 subjects were randomized to 1 of the following 4 sequential ascending REGN2477 IV dose panels where 8 subjects were randomized in a 6:2 ratio into each of the first 3 Panels (Panels A, B, and C) and 24 subjects were randomized in a 1:1:1:1 ratio (6 subjects each) into Panel D.

- Panel A: 4 subjects REGN1033 (6 mg/kg IV) + REGN2477 low dose (1 mg/kg IV) or 2 subjects placebo
- Panel B: 4 subjects REGN1033 (6 mg/kg IV) + REGN2477 medium dose (3 mg/kg IV) or 2 subjects placebo
- Panel C: 4 subjects REGN1033 (6 mg/kg IV) + REGN2477 high dose (10 mg/kg IV) or 2 subjects placebo
- Panel D: REGN1033(6 mg/kg IV + REGN2477 high dose (10 mg/kg IV) placebo, REGN2477 (10 mg/kg IV), or REGN1033 (6 mg/kg IV)

[00222] Subjects received a single intravenous dose of one or both of anti-GDF8 antibody REGN1033 and/or anti-Activin A antibody REGN2477. In the primary analyses, the placebo and high dose combination groups were pooled across panels, yielding 12 subjects on placebo and 12 on the high dose combination, as shown in Figure 2A. The dosing schedule shown in Figure 2A was used in studies shown in each of the subsequent Figures 2B to 17.

[00223] Subjects participated in a screening period of up to 28 days, followed by a baseline and treatment visit on day 1, and a follow-up period of 113 days.

Efficacy and Safety Analysis

[00224] The full analysis set (FAS) includes all randomized subjects; it is based on the treatment allocated (as randomized). Efficacy endpoints were analyzed using the FAS. The safety analysis set (SAF) includes all randomized subjects who received any study drug; it is based on the treatment received (as treated). Treatment compliance/administration and all clinical safety variables were analyzed using the SAF.

[00225] The efficacy variables included: Thigh muscle tissue volume, excluding and including intramuscular adipose tissue and large vessels as measured by Magnetic Resonance Imaging (MRI);Total lean mass as measured by dual X-ray absorptiometry

(DXA); Appendicular lean body mass (calculated by a LBM equation) as measured by DXA; and Total fat mass as measured by DXA.

[00226] The demographics and baseline characteristics of the subjects were balanced across the treatment groups, as per the below Table 5:

[00227] Table 5. Baseline Characteristics were balanced across treatment groups

	Placebo	Anti-GDF8	Anti-Activin A (10 mg/kg) IV	Anti-GDF8 + anti-activin A (1 mg/kg) IV	Anti-GDF8 + anti-activin A (3 mg/kg) IV	Anti-GDF8 + anti-activin A (10 mg/kg) IV
	(N=12)	(N=6)	(N=6)	(N=6)	(N=6)	(N=12)
Age (Years)						
Median	54	56	61	60	60	55.5
Height (cm)						
Median	162.5	164.2	162.5	166.5	165	163.5
Weight (kg)						
Median	68.35	70.5	69.6	61.35	67.05	70.4
BMI (kg/m^2)						
Median	26.2	26.55	26.65	22.9	25.55	26.6

Statistical methods

[00228] The percent change and change of efficacy variables from baseline to either week 4 or 8 in the full analysis set (FAS) were analyzed using analysis of covariance (ANCOVA) model with treatment group as fixed effect, and the baseline value as continuous covariate. Least-squares means at week 4 and week 8 for each treatment group with the corresponding standard error, confidence interval, and the p value for treatment comparisons were provided from this model. Placebo subjects were pooled across panels. Missing efficacy data was not imputed. No adjustment for multiple testing is applied in this study.

RESULTS

[00229] A total of 48 subjects were randomized, administered study drug and completed the study. One subject in the REGN2477+REGN1033 high dose group had an interrupted infusion of study drug due to an adverse event of 'Infusion site swelling'.

Efficacy

[00230] The blockade of both Activin A and GDF8 in combination increased thigh muscle volume and decreased fat mass at week 8, as shown in Figures 2B and 2C. The greatest effect was seen at the highest dose combination.

[00231] Figure 2B shows a bar graph showing % change in thigh muscle volume by MRI at 8 weeks after a single dose in postmenopausal women. Numbers show changes from placebo. * indicates nominal $p<0.05$ vs. placebo; **** indicates nominal $p< 0.0001$ vs. placebo. The single dose amounts as mg/kg of either anti-Activin A antibody and/or anti-GDF8 antibody are shown below the bar graph. Treatment with REGN2477 + REGN1033 in medium and high dose combinations resulted in significantly increased thigh muscle volume ($p < 0.05$; $p<0.0001$, respectively) in a dose-related manner compared with placebo. Subjects in high dose panel exhibited up to 7.73% as % change from baseline versus 0.88% with placebo at week 8.

[00232] Figure 2C shows a bar graph showing % change in total fat mass by DXA at 8 weeks after a single dose in postmenopausal women. Numbers show changes from placebo. * indicates nominal $p<0.05$ vs. placebo; **** indicates nominal $p< 0.0001$ vs. placebo. The single dose amounts as mg/kg of either anti-Activin A antibody and/or anti-GDF8 antibody are shown below the bar graph. Treatment with high dose REGN2477 + REGN1033 combination resulted in significantly decreased total fat mass ($p<0.05$) compared with placebo.

[00233] As shown in Figure 3, anti-Activin A antibody REGN2477 combined with anti-GDF8 antibody REGN1033 led to dose-dependent increases in thigh muscle volume. The thigh muscle volume results, as measured via MRI, excluding intermuscular adipose tissue and large vessels, are likewise summarized in Table 6, below. Treatment with mid- and high dose REGN2477 + REGN1033 combination resulted in significantly increased thigh muscle volume ($p<0.05$) compared with placebo, as shown in Table 6.

[00234] Table 6. Thigh Muscle Volume measured by MRI*

		Placebo	anti-GDF8	anti-activin A High Dose	anti-GDF8 + anti-activin A Low Dose	anti-GDF8 + anti-activin A Mid Dose	anti-GDF8 + anti-activin A High Dose
Week 4, % change	N	12	6	6	6	6	12
	mean	0.4	3.3	0.3	4.9	4.6	5.9
Week 8, % change	mean	0.9	4.6	2.9	3.4	6.2	7.8
	□ LSM vs placebo (SE)		3.73 (1.819)	1.97 (1.821)	2.63 (1.824)	5.31 (1.817)	6.85 (1.484)
	Nominal p-value		0.0467	0.2846	0.1569	0.0056	<0.0001

*excluding intermuscular adipose tissue and large vessels

[00235] As shown in Table 4, treatment with REGN2477 + REGN1033 in medium and high dose combinations resulted in significantly increased thigh muscle volume in a dose-related manner compared with placebo.

[00236] Figure 4 shows that increases in thigh muscle volume were consistently observed in individual subjects following treatment with anti-Activin A + anti-GDF8 in combination. Within each treatment group, different lines indicate different individuals.

[00237] The pattern of effects on appendicular lean mass (the sum of lean mass of arms and legs) with anti-Activin A antibody REGN2477+ anti-GDF8 antibody REGN1033 in combination was similar to that seen on thigh muscle volume, as shown in Figure 5. The appendicular lean mass results, as measured via DXA, are likewise summarized in Table 7. The mid and high dosing combinations significantly increased appendicular lean mass compared to placebo, as shown in Table 7.

[00238] Table 7. Appendicular Lean Mass by DXA

		Placebo	Anti-GDF8	Anti-activin A High Dose	Anti-GDF8 + anti-activin A Low Dose	Anti-GDF8 + anti-activin A Mid Dose	Anti-GDF8 + anti-activin A High Dose
	N	12	6	6	6	6	12
Baseline, kg	Mean	17.44	18.10	18.02	17.92	18.67	17.32
Week 4, % Change	Mean	-0.2	1.2	2.1	4.2	4.6	3.8
Week 8, % Change	Mean	0.8	2.6	2.3	3.7	6.9	5.8
	ΔLSM vs Placebo (SE)		1.90 (1.266)	1.61 (1.265)	2.96 (1.264)	6.39 (1.275)	4.97 (1.031)
	Nominal p-value		0.1418	0.2109	0.0242	<0.0001	<0.0001

[00239] As shown in Figure 6, blockade of both Activin A and GDF8 led to reductions in total fat mass, as assessed by DXA. The high dose combination of anti-GDF8 antibody REGN1033 and anti-Activin A antibody REGN2477 significantly reduced total fat mass by DXA compared to placebo at week 8 (*p <0.05). The total fat mass results, as measured via DXA, are likewise summarized in Table 8, below.

[00240] Table 8. Total Fat Mass by DXA

		anti-GDF8	anti-activin A	anti-GDF8 + anti-activin A	anti-GDF8 + anti-activin A	anti-GDF8 + anti-activin A
	Dose	High	High	Low	Mid	High
Week 8, % Change	N	6	6	6	6	12
	ΔLSM vs Placebo (SE)	-2.11 (1.96)	-0.69 (1.93)	-0.2 (1.93)	-2.08 (1.93)	-3.92 (1.58)*

* p<0.05

[00241] Blockade of Activin A and GDF8 was also found to be associated with decreases in android fat mass, as assessed by DXA, as shown in Figure 7. The high dose combination of anti-GDF8 antibody REGN1033 and anti-Activin A antibody REGN2477

significantly reduced android fat mass by DXA compared to placebo at week 8 (*p <0.05).

[00242] Further efficacy results are shown in Figures 8 to 17 and summarized in the tables, below. REGN2477 +REGN1033 high, medium and low doses are shown in Figure 2A.

[00243] Compared with placebo, REGN2477 +REGN1033 medium ($p < 0.05$) and high dose groups ($p < 0.001$) exhibited significantly increased thigh muscle volume, excluding Intramuscular adipose tissue and large vessels (Figure 8). Thigh muscle volume increased in the REGN2477 + REGN1033 high dose group by 7.73% as compared with 0.88% in the placebo group (nominal $p<0.001$) at 8 weeks. Compared with placebo, REGN2477 + REGN1033 at the medium dose and REGN1033 alone also significantly increased thigh muscle volume. Increases in thigh muscle volume were consistently observed in individual subjects treated with the combination in a dose responsive manner. (data not shown).

[00244] Compared with placebo, REGN2477 +REGN1033 high dose group exhibited significantly increased total lean mass by DXA ($p < 0.05$) (Figure 9).

[00245] Appendicular lean body mass (calculated via aLBM equation), was significantly increased in each of the combination REGN2477 + REGN1033 treatment groups compared with placebo (low dose $p<0.05$, medium and high dose groups $p < 0.001$)(Figure 10). Appendicular lean body mass increased in the REGN2477 + REGN1033 medium dose group by 7.15% from baseline as compared with 0.76% in the placebo group at 8 weeks. Similarly, appendicular lean body mass increased in the REGN2477 + REGN1033 high dose group by 5.7% from baseline as compared with 0.76% in the placebo group.

[00246] Total fat mass was significantly decreased in the high dose REGN2477 + REGN1033 treatment group; total fat mass was decreased: 3.92%(high dose group) compared with placebo at 0.5%(nominal $p<0.05$)(Figure 11).

[00247] Compared to placebo, thigh muscle volume including intramuscular adipose tissue and large vessels was significantly increased in each of the medium and high dose REGN2477 +REGN1033 groups and REGN1033 group at 4 weeks, and at 8 weeks (Figure 12). Low dose REGN2477+REGN1033 also exhibited significant increase in thigh muscle volume including intramuscular adipose tissue and large vessels compared to placebo at 4 weeks.

[00248] Appendicular lean mass (sum of arms and legs) was significantly increased in each of combination REGN2477 + REGN1033 treatment groups ($p<0.05$) at 4 and 8 weeks compared to placebo (Figure 13).

[00249] Android fat mass in the high dose REGN2477 + REGN1033 treatment group was also significantly reduced (Figure 14). Android fat mass was reduced 6.6% in the high dose REGN2477+REGN1033 group as compared to no reduction in the placebo.

[00250] Thigh intramuscular adipose tissue volume (cm3) was significantly increased in the high dose REGN2477+REGN1033 group ($p<0.05$) as compared to placebo at 8 weeks (Figure 15).

[00251] Reductions in adipose tissue in the high dose group were observed in the sum of Intramuscular and Perimuscular Adipose Tissue (IMAT)(Figure 16) and in the low and medium dose group of subcutaneous adipose tissue. (Figure 17). In contrast, thigh intramuscular adipose tissue was increased in the high dose group to 8% as compared with placebo with a reduction of 4%. (Figure 17).

[00252] REGN2477+REGN1033, in the high dose group, significantly increased all of the major measures of muscle volume and lean mass compared with placebo, at both Week 4 and Week 8; the effects at Week 4 were generally less pronounced than at Week 8 (Table 9). A summary of Percent Change in Key Body Composition Measures at Week 8 (Full Analysis Set, LS Means and SE presented) is shown in Table 9, below.

[00253] Table 9. Summary of Change in Body Composition at Week 8

Endpoint					Low	Medium	High
		Placebo	R1033 (6 mg/kg)	R2477 (10 mg/kg)	R2477 (1 mg/kg)	R2477 (3 mg/kg)	R2477 (10 mg/kg)
					+R1033	+R1033	+R1033
		(n= 12)	(n= 6)	(n= 6)	(n= 6)	(n= 6)	(n= 12)
Thigh Muscle Volume	% Change from baseline	0.88 (1.05)	4.61 (1.49)	2.85 (1.49)	3.51 (1.49)	6.19 (1.48)	7.73 (1.05)
	Difference from placebo		3.73 (1.82)*	1.97 (1.82)	2.63 (1.82)	5.31 (1.82)*	6.85 (1.48)**
Total Lean Mass by DXA	% Change from baseline	1.31 (0.64)	2.18 (0.89)	1.94 (0.90)	3.16 (0.89)	4.67 (0.89)	4.31 (0.63)
	Difference		0.88	0.63	1.85	3.37	3.00

	from placebo		(1.10)	(1.11)	(1.10)	(1.10)*	(0.89)*
Appen-dicular Lean Body Mass	% Change from baseline	0.76 (0.73)	2.65 (1.03)	2.37 (1.03)	3.72 (1.03)	7.15 (1.04)	5.72 (0.73)
	Difference from placebo		1.90 (1.27)	1.61 (1.26)	2.96 (1.26)*	6.39 (1.27)**	4.97 (1.03)**
Total Fat Mass by DXA	% Change from baseline	-0.65 (1.12)	-2.76 (1.60)	-1.34 (1.58)	-0.85 (1.58)	-2.73 (1.58)	-4.57 (1.12)
	Difference from placebo		-2.11 (1.96)	-0.69 (1.93)	-0.2 (1.93)	-2.08 (1.93)	-3.92 (1.58)*

* $p < 0.05$; ** $p < 0.001$

[00254] In Table 9, above, changes from baseline and differences from placebo are Least-Squares (LS) means based on the ANCOVA model with baseline as a covariate and treatment as a fixed factor. Standard errors (SE) and p-values also taken from the ANCOVA. Nominal p-values are reported.

[00255] Bone mineral density (BMD) mass and bone mineral content (BMC) mass were measured by DXA, as shown in Table 10. At the high dose, R2477 + R1033 increased Bone Mineral Content as measured by DXA, while total bone mineral density did not change (Table 10).

[00256] Sum of fat mass of arms and legs were measured as shown in Table 10. At the high dose, R2477 + R1033 decreased sum of fat mass of arms and legs ($p< 0.05$).

[00257] The data from the primary endpoint analysis is shown in Table 10, below.

[00258] Table 10: Primary analysis of efficacy endpoints

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
Thigh Muscle Tissue Volume (cm3), Excluding Intra-muscular Adipose Tissue and Large Vessels/% change	LS Mean (SE)Week 4	0.37 (0.76)	3.27 (1.07)	0.27 (1.08)	4.96 (1.08)	4.61 (1.07)	5.92 (0.76)
	LS Mean Diff (SE)/ week 4		2.9 (1.32) *	-0.11 (1.32)	4.59 (1.32)*	4.24 (1.31)*	5.55 (1.07)**
	LS Mean (SE)/ week 8	0.88 (1.05)	4.61(1.49)	2.85 (1.49)	3.51 (1.49)	6.19 (1.48)	7.73 (1.05)
	LS Mean Diff (SE)/ week 8		3.73 (1.82) *	1.97 (1.82)	2.63 (1.82)	5.31 (1.82)*	6.85 (1.48)**
Change	LS Mean (SE)/week 4	1.64 (3.26)	13.95 (4.61)	-0.14 (4.63)	20.29 (4.63)	18.77 (4.61)	24.5 (3.26)
	LS Mean Diff (SE)/week 4		12.32 (5.65) *	-1.78 (5.66)	18.65 (5.67)*	17.14 (5.64)*	22.86 (4.61)**

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean (SE)/week 8	3.5 (4.13)	20.42 (5.85)	11.22 (5.87)	14.55 (5.88)	25.44 (5.85)	31.95 (4.14)
	LS Mean Diff (SE)/ week 8		16.91 (7.17) *	7.71 (7.18)	11.05 (7.19)	21.94 (7.16)*	28.45 (5.85)**
Thigh Muscle Volume (cm3), Including Intra-muscular Adipose Tissue and Large Vessels/% change	LS Mean (SE)/ week 4	0.21 (0.78)	3.32 (1.1)	-0.2 (1.11)	5 (1.11)	4.49 (1.1)	5.76 (0.78)
	LS Mean Diff (SE)/ week 4		3.1 (1.35) *	-0.41 (1.36)	4.79 (1.35)*	4.28 (1.35)*	5.54 (1.1)**
	LS Mean (SE)/ week 8	0.85 (1.06)	4.14 (1.51)	2.47 (1.52)	3.14 (1.51)	5.99 (1.5)	7.41 (1.06)
	LS Mean Diff (SE)/ week 8		3.29 (1.84)	1.62 (1.85)	2.29 (1.85)	5.14 (1.84)*	6.57 (1.5)**
change	LS Mean (SE)/ week 4	0.99 (3.45)	14.76 (4.89)	-2.61 (4.94)	21.22 (4.91)	19.35 (4.89)	25.22 (3.45)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 4		13.77 (5.99) *	-3.6 (6.02)	20.24 (6.01)*	18.36 (5.98)*	24.23 (4.88)**
	LS Mean (SE)/week 8	3.55 (4.42)	19.54 (6.25)	9.26 (6.32)	13.64 (6.29)	26.04 (6.25)	32.27 (4.42)
	LS Mean Diff (SE)/week 8		15.99 (7.66) *	5.72 (7.7)	10.09 (7.69)	22.49 (7.65)*	28.72 (6.25)**
Appen-dicular Lean Body Mass (Calcu-lated by aLBM Equation) (kg)/ % change	LS Mean (SE)/ week 4	-0.31 (0.74)	1.26 (1.04)	2.15 (1.04)	4.26 (1.04)	4.77 (1.05)	3.7 (0.74)
	LS Mean Diff (SE)/ week 4		1.57 (1.28)	2.46 (1.28)	4.57 (1.28)**	5.08 (1.29)**	4.01 (1.04)**
	LS Mean (SE)/week 8	0.76 (0.73)	2.65 (1.03)	2.37 (1.03)	3.72 (1.03)	7.15 (1.04)	5.72 (0.73)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 8		1.9 (1.27)	1.61 (1.26)	2.96 (1.26)*	6.39 (1.27)**	4.97 (1.03)**
Change	LS Mean (SE)/ week 4	-0.05 (0.13)	0.23 (0.18)	0.35 (0.18)	0.73 (0.18)	0.84 (0.18)	0.63 (0.13)
	LS Mean Diff (SE)/ week 4		0.28 (0.22)	0.4 (0.22)	0.78 (0.22)*	0.89 (0.22)**	0.68 (0.18)**
	LS Mean (SE)	0.11 (0.13)	0.45 (0.19)	0.41 (0.19)	0.64 (0.18)	1.28 (0.19)	1.02 (0.13)
	LS Mean Diff (SE)/week 8		0.33(0 .23)	0.29 (0.23)	0.53 (0.23)*	1.16 (0.23)**	0.91 (0.18)**
Total Lean Mass (kg)/ % change	LS Mean (SE)/week 4	0.16 (0.56)	0.71 (0.79)	1.53 (0.79)	3.42(0.79)	3.52 (0.79)	2.84 (0.56)
	LS Mean Diff (SE)/week 4		0.55 (0.97)	1.37 (0.98)	3.26 (0.98)*	3.36 (0.98)*	2.69 (0.79)*
	LS Mean (SE)/week 8	1.31 (0.64)	2.18 (0.89)	1.94 (0.9)	3.16 (0.89)	4.67 (0.89)	4.31 (0.63)
	LS Mean Diff (SE)/week 8		0.88 (1.1)	0.63 (1.11)	1.85 (1.1)	3.37 (1.1)*	3 (0.89)*

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
change	LS Mean (SE)/week 4	0.08 (0.23)	0.25 (0.33)	0.61 (0.33)	1.42 (0.33)	1.44 (0.33)	1.14 (0.23)
	LS Mean Diff (SE)/week 4		0.17 (0.41)	0.54 (0.41)	1.34 (0.41)*	1.36 (0.41)*	1.07 (0.33)*
	LS Mean (SE)/week 8	0.53 (0.27)	0.86 (0.37)	0.78 (0.37)	1.27 (0.37)	1.92 (0.37)	1.8 (0.26)
	LS Mean Diff (SE)/week 8		0.33 (0.46)	0.25 (0.46)	0.74 (0.46)	1.39 (0.46)*	1.26 (0.37)*
Appen-dicular Lean Mass (Sum of Lean Mass of Arms and Legs) (kg)/ % change	LS Mean (SE)/week 4	-0.35 (0.74)	1.2 (1)	2.02 (1)	4.06 (1)	4.51 (1)	3.54 (0.71)
	LS Mean Diff (SE)/week 4		1.56 (1.25)	2.37 (1.24)	4.42 (1.24)**	4.87 (1.26)**	3.89 (1.02)**

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean (SE)/week 8	0.95 (0.72)	2.51 (0.97)	2.22 (0.97)	3.53 (0.97)	6.74 (0.98)	5.48 (0.69)
	LS Mean Diff (SE)/week 8		1.56 (1.22)	1.28 (1.21)	2.58 (1.21)*	5.79 (1.23)**	4.53 (0.99)**
change	LS Mean (SE)/week 4	-0.06 (0.14)	0.23 (0.18)	0.35 (0.18)	0.73 (0.18)	0.83 (0.19)	0.64 (0.13)
	LS Mean Diff (SE)/week 4		0.28 (0.23)	0.41 (0.23)	0.79 (0.23)*	0.89 (0.23)**	0.69 (0.19)**
	LS Mean (SE)/week 8	0.17 (0.14)	0.44 (0.18)	0.4 (0.18)	0.64 (0.18)	1.26 (0.18)	1.02 (0.13)
	LS Mean Diff (SE)/week 8		0.27 (0.23)	0.23 (0.23)	0.48 (0.23)*	1.09 (0.23)**	0.86 (0.19)**
Total Fat Mass (kg)/% change	LS Mean (SE)/week 4	-0.11 (0.81)	-1.15 (1.15)	1.32 (1.14)	-0.76 (1.14)	-1.52 (1.14)	-2.58 (0.8)
	LS Mean Diff (SE)/week 4		-1.04 (1.41)	1.42 (1.39)	-0.65 (1.39)	-1.41 (1.39)	-2.47 (1.14)*

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean (SE)/week 8	-0.65 (1.12)	-2.76 (1.6)	-1.34 (1.58)	-0.85 (1.58)	-2.73 (1.58)	-4.57 (1.12)
	LS Mean Diff (SE)/week 8		-2.11 (1.96)	-0.69 (1.93)	-0.2 (1.93)	-2.08 (1.93)	-3.92 (1.58)*
change	LS Mean (SE)/week 4	0.07 (0.21)	-0.18 (0.3)	0.29 (0.29)	-0.29 (0.29)	-0.38 (0.29)	-0.6 (0.21)
	LS Mean Diff (SE)/ week 4		-0.25 (0.36)	0.23 (0.36)	-0.36 (0.36)	-0.45 (0.36)	-0.67 (0.29)*
	LS Mean (SE)/week 8	-0.04 (0.28)	-0.54 (0.4)	-0.46 (0.4)	-0.34 (0.4)	-0.75 (0.4)	-1.16 (0.28)
	LS Mean Diff (SE)/week 8		-0.5 (0.49)	-0.42 (0.49)	-0.3 (0.49)	-0.71 (0.49)	-1.12 (0.4)*
Android Fat Mass (kg)/ % change	LS Mean (SE)/week 4	-0.21 (1.3)	-1.57 (1.85)	0.44 (1.84)	-2.47 (1.84)	-1.8 (1.83)	-3.94 (1.3)
	LS Mean Diff (SE)/week 4		-1.35 (2.26)	0.65 (2.25)	-2.25 (2.25)	-1.59 (2.25)	-3.73 (1.83)*
	LS Mean (SE)	-0.05 (1.69)	-2.35 (2.4)	-4.67 (2.39)	-1.71 (2.39)	-2.62 (2.38)	-6.65 (1.69)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 8		-2.31 (2.93)	-4.62 (2.92)	-1.66 (2.93)	-2.57 (2.92)	-6.61 (2.38)*
change	LS Mean (SE)/week 4	0.03 (0.03)	0.01 (0.04)	0 (0.04)	-0.05 (0.04)	-0.04 (0.04)	-0.07 (0.03)
	LS Mean Diff (SE)/week 4		-0.01 (0.05)	-0.02 (0.05)	-0.07 (0.05)	-0.06 (0.05)	-0.1 (0.04)*
	LS Mean (SE)/week 8	0.03 (0.04)	-0.01 (0.05)	-0.09 (0.05)	-0.03 (0.05)	-0.07 (0.05)	-0.13 (0.04)
	LS Mean Diff (SE)/week 8		-0.04 (0.06)	-0.11 (0.06)	-0.06 (0.06)	-0.1 (0.06)	-0.16 (0.05)*
Thigh Intra-muscular Adipose Tissue Volume (cm3)/ % volume	LS Mean (SE)/week 4	-2.9 (3)	-2 (4.24)	-3.44 (4.23)	6.19 (4.28)	-2.99 (4.24)	8.25 (2.98)
	LS Mean Diff (SE)/week 4		0.9 (5.22)	-0.54 (5.17)	9.09 (5.27)	-0.09 (5.17)	11.15 (4.23)*

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean (SE)/week 8	-4.19 (4.21)	-3.35 (5.96)	0.65 (5.95)	1.14 (6.02)	-3.88 (5.96)	7.26 (4.19)
	LS Mean Diff (SE)/week 8		0.84 (7.34)	4.84 (7.26)	5.33 (7.41)	0.31 (7.27)	11.45 (5.95)
change	LS Mean (SE)/week 4	-0.09 (0.13)	-0.07 (0.18)	-0.2 (0.18)	0.14 (0.18)	-0.12 (0.18)	0.24 (0.13)
	LS Mean Diff (SE)/week 4		0.02 (0.22)	-0.11 (0.22)	0.23 (0.22)	-0.03 (0.22)	0.33 (0.18)
	LS Mean (SE)/week 8	-0.17 (0.17)	-0.13 (0.24)	-0.02 (0.24)	-0.08 (0.25)	-0.22 (0.24)	0.2 (0.17)
	LS Mean Diff (SE)/week 8		0.04 (0.3)	0.15 (0.3)	0.09 (0.3)	-0.05 (0.3)	0.37 (0.24)
Sum of Intra-muscular and Peri-muscular Adipose Tissue (IMAT)/ % change	LS Mean (SE)/week 4	0.98 (1.4)	-3.09 (1.98)	1.63 (1.99)	-2.09 (2.03)	1.75 (1.99)	-1.54 (1.4)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 4		-4.07 (2.43)	0.65 (2.43)	-3.07 (2.47)	0.77 (2.43)	-2.52 (1.98)
	LS Mean (SE)/week 8	0.94 (1.49)	-0.12 (2.12)	0.26 (2.13)	-0.5 (2.17)	-1.63 (2.12)	-2.76 (1.49)
	LS Mean Diff (SE)/week 8		-1.06 (2.6)	-0.68 (2.59)	-1.44 (2.64)	-2.57 (2.59)	-3.7 (2.11)
Change	LS Mean (SE)/week 4	1.05 (1.65)	-3.45 (2.34)	2.29 (2.35)	-3.2 (2.4)	1.93 (2.35)	-1.75 (1.65)
	LS Mean Diff (SE)/week 4		-4.5 (2.87)	1.24 (2.87)	-4.25 (2.92)	0.88 (2.87)	-2.8 (2.33)
	LS Mean (SE)/week 8	0.69 (1.8)	0.28 (2.56)	0.48 (2.57)	-1.73 (2.62)	-1.97 (2.57)	-3.04 (1.81)
	LS Mean Diff (SE)/week 8		-0.4 (3.14)	-0.21 (3.13)	-2.42 (3.19)	-2.66 (3.13)	-3.73 (2.55)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
Sub-cutaneous Adipose Tissue Volume (cm3)/ % change	LS Mean (SE)/week 4	-1.27 (1.02)	0.05 (1.45)	-0.26 (1.47)	-1.67 (1.44)	-1.98 (1.44)	-1.76 (1.02)
	LS Mean Diff (SE)/week 4		1.32 (1.78)	1.01 (1.82)	-0.4 (1.76)	-0.71 (1.76)	-0.49 (1.44)
	LS Mean (SE)/week 8	1.31 (1.21)	1.01 (1.71)	1.56 (1.75)	-3.73 (1.71)	-3.04 (1.7)	-1.45 (1.21)
	LS Mean Diff (SE)/week 8		-0.31 (2.11)	0.25 (2.15)	-5.04 (2.09)*	-4.36 (2.09)*	-2.77 (1.7)
change	LS Mean (SE)/week 4	-3.94 (3.83)	0.35 (5.42)	-0.74 (5.52)	-8.23 (5.39)	-3.02 (5.39)	-7.29 (3.82)
	LS Mean Diff (SE)/week 4		4.29 (6.68)	3.19 (6.8)	-4.3 (6.6)	0.92 (6.6)	-3.35 (5.39)
	LS Mean (SE)/week 8	4.74 (4.74)	5.7 (6.7)	5.53 (6.83)	-17.58 (6.67)	-8.75 (6.66)	-5.55 (4.73)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 8		0.95 (8.26)	0.79 (8.41)	-22.33 (8.16)*	-13.5 (8.17)	-10.3 (6.66)
Sum of Fat Mass of Arms and Legs (kg)/ % change	LS Mean (SE)/week 4	-0.8 (1.01)	-0.59 (1.44)	1.89 (1.42)	0.48 (1.42)	-2.87 (1.42)	-3.31 (1)
	LS Mean Diff (SE)/week 4		0.21 (1.78)	2.69 (1.74)	1.28 (1.74)	-2.07 (1.74)	-2.51 (1.42)
	LS Mean (SE)/week 8	-0.47 (1.34)	-2.44 (1.9)	0.11 (1.87)	0.27 (1.87)	-3.51 (1.87)	-5.2 (1.32)
	LS Mean Diff (SE)/week 8		-1.97 (2.35)	0.58 (2.31)	0.74 (2.3)	-3.04 (2.3)	-4.73 (1.88)*
change	LS Mean (SE)/week 4	-0.09 (0.12)	-0.06 (0.17)	0.21 (0.16)	0.01 (0.16)	-0.33 (0.16)	-0.41 (0.11)
	LS Mean Diff (SE)/week 4		0.02 (0.2)	0.3 (0.2)	0.09 (0.2)	-0.25 (0.2)	-0.33 (0.16)
	LS Mean (SE)/week 8	-0.05 (0.15)	-0.26 (0.22)	-0.03 (0.21)	-0.07 (0.21)	-0.41 (0.21)	-0.62 (0.15)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 8		-0.21 (0.27)	0.02 (0.26)	-0.02 (0.26)	-0.37 (0.26)	-0.58 (0.22)*
Total Bone Mineral Density (BMD) Mass (g/cm2)/% change	LS Mean (SE)/week 4	0.05 (0.25)	-0.22 (0.36)	-0.48 (0.35)	-0.04 (0.36)	0.47 (0.36)	0.55 (0.25)
	LS Mean Diff (SE)/week 4		-0.27 (0.43)	-0.53 (0.43)	-0.09 (0.44)	0.42 (0.44)	0.5 (0.35)
	LS Mean (SE)/week 8	-0.01 (0.28)	-0.47 (0.4)	-0.72 (0.4)	-0.08 (0.4)	-0.22 (0.41)	0.16 (0.28)
	LS Mean Diff (SE)/week 8		-0.46 (0.49)	-0.71 (0.49)	-0.07 (0.5)	-0.21 (0.5)	0.17 (0.4)
change	LS Mean (SE)/week 4	0 (0.003)	-0.002 (0.004)	-0.005 (0.004)	0 (0.004)	0.005 (0.004)	0.006 (0.003)
	LS Mean Diff (SE)/week 4		0 (0)	-0.01 (0)	0 (0)	0 (0)	0.01 (0)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean (SE)/week 8	0 (0.003)	-0.006 (0.004)	-0.008 (0.004)	0 (0.004)	-0.002 (0.004)	0.002 (0.003)
	LS Mean Diff (SE)/week 8		-0.01 (0.01)	-0.01 (0.01)	0 (0.01)	0 (0.01)	0 (0)
Total Bone Mineral Content (BMC) Mass (kg)/ % change	LS Mean (SE)/week 4	0.1 (0.28)	-0.16 (0.4)	-0.23 (0.4)	0.25 (0.4)	-0.19 (0.4)	0.62 (0.28)
	LS Mean Diff (SE)/week 4		-0.26 (0.49)	-0.33 (0.49)	0.15 (0.49)	-0.29 (0.49)	0.52 (0.4)
	LS Mean (SE)/week 8	-0.42 (0.26)	-0.27 (0.36)	-0.48 (0.36)	-0.08 (0.36)	-0.56 (0.36)	0.65 (0.25)
	LS Mean Diff (SE)/week 8		0.16 (0.45)	-0.05 (0.44)	0.34 (0.44)	-0.14 (0.44)	1.07 (0.36)*
change	LS Mean (SE)/week 4	0.002 (0.006)	-0.003 (0.009)	-0.005 (0.009)	0.006 (0.009)	-0.003 (0.009)	0.014 (0.006)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 4		-0.01 (0.01)	-0.01 (0.01)	0 (0.01)	-0.01 (0.01)	0.01 (0.01)
	LS Mean (SE)/week 8	-0.009 (0.006)	-0.007 (0.008)	-0.011 (0.008)	-0.001 (0.008)	-0.012 (0.008)	0.014 (0.006)
	LS Mean Diff (SE)/week 8		0 (0.01)	0 (0.01)	0.01 (0.01)	0 (0.01)	0.02 (0.01)*
T-score for Total Body/ % change	LS Mean (SE)/week 4	-18.26 (18.01)	-51.19 (25.74)	-2.95 (25.45)	-6.66 (25.7)	-39.76 (25.87)	-0.9 (17.96)
	LS Mean Diff (SE)/week 4		-32.93 (31.23)	15.31 (31.1)	11.6 (31.56)	-21.5 (31.73)	17.37 (25.43)
	LS Mean (SE)/week 8	-20.87 (17.94)	-58.54 (25.64)	3.83 (25.35)	5.16 (25.6)	23.12 (25.77)	8.49 (17.89)
	LS Mean Diff (SE)/week 8		-37.67 (31.11)	24.71 (30.98)	26.04 (31.44)	44 (31.61)	29.36 (25.33)
change	LS Mean (SE)/week 4	0.01 (0.03)	-0.03 (0.05)	-0.07 (0.05)	0 (0.05)	0.06 (0.05)	0.08 (0.03)

Parameter	Analysis Visit	Placebo (n= 12)	R1033 (n= 6)	R2477 (10 mg/kg) IV (n= 6)	R1033 +R2477 (1 mg/kg) IV (n= 6)	R1033 +R2477 (3 mg/kg) IV (n= 6)	R1033 +R2477 (10 mg/kg) IV (n= 12)
	LS Mean Diff (SE)/week 4		-0.04 (0.06)	-0.07 (0.06)	-0.01 (0.06)	0.06 (0.06)	0.07 (0.05)
	LS Mean (SE)/week 8	0 (0.04)	-0.07 (0.05)	-0.1 (0.05)	-0.01 (0.05)	-0.03 (0.06)	0.03 (0.04)
	LS Mean Diff (SE)/week 8		-0.07 (0.07)	-0.1 (0.07)	-0.01 (0.07)	-0.03 (0.07)	0.03 (0.05)

Note: Least-squares (LS) means, standard errors (SE) and p-value taken from ANCOVA. The model includes baseline measurement as covariate and the treatment as fixed factor. * indicated p-value < 0.05; ** indicated p-value < 0.001.

[00259] Compared with placebo, R2477+R1033 combination significantly increased thigh muscle volume and total lean mass in medium and high dose groups (Figures 8 and 9), significantly increased appendicular lean body mass in all dose groups (Figure 10) and significantly decreased total fat mass as well as android fat mass in the high dose group (Table 10). R2477 + R1033, in the high dose group, significantly increased all of the major measures of muscle volume and lean mass compared with placebo, at both Week 4 and Week 8; the effects at Week 4 were generally less pronounced than at Week 8 (Table 10). At Week 8, the high dose anti-Activin A R2477 + anti-GDF8 R1033 group exhibited increased % change in total bone mineral content from placebo, as measured by DXA, while total bone mineral density did not change (Table 10).

SAFETY

[00260] All treatment emergent adverse events (TEAEs) were mild to moderate in severity except one severe TEAE of 'radius fracture' reported by a placebo subject. There were no serious adverse events, no deaths, and no discontinuations due to TEAEs. Headache was the most frequent TEAE in each of the treatment groups, occurring in 58.3% of all study subjects and in 50% of placebo subjects. Muscle spasms, nausea and

mouth ulceration were the other frequent TEAEs in REGN2477+REGN1033 groups that occurred in 25% or more of subjects in the combination R2477 +R1033 groups; these TEAEs occurred less frequently in the placebo group, but there does not appear to be any clear dose-response relationship. There were no clear signals of bleeding or diarrhea, adverse events that have been associated with blockade of activin receptors. One TEAE of a nosebleed (preferred term of epistaxis) occurred in a R2477 + R1033 dose group – it resolved after 9 minutes.

[00261] Review of Potentially Clinically Significant Values (PCSVs) revealed no significant differences between REGN2477+REGN1033 and Placebo in Labs, Vital signs, and ECG that would indicate negative effects of REGN2477+REGN1033. Within laboratory, vital sign and ECG categories, there were 0-2 subjects with PCSVs in the combined REGN2477+REGN1033 dose groups (N of 24); however the percentage of subjects with PCSVs was equal to or lower than that found in the placebo group. There were no treatment-emergent PCSVs related to liver function tests.

Conclusions

[00262] In healthy postmenopausal women, single intravenous doses of REGN2477 + REGN1033 increased thigh muscle volume, total lean mass, and appendicular lean body mass. One surprising finding was the uniformity of the thigh muscle changes: all of the individuals exposed to the combination exhibited an increase in thigh muscle volume, as shown in Figure 4. In addition, single intravenous doses of REGN2477+REGN1033 decreased total fat, and in particular, android fat mass. Treatment with REGN1033 alone increased thigh muscle volume.

[00263] In general, REGN2477, REGN1033, and REGN2477+REGN1033 in this clinical study were considered to have an acceptable safety profile and were well tolerated. There were no serious adverse events.

[00264] The present invention is not to be limited in scope by the specific embodiments described herein. Indeed, various modifications of the invention in addition to those described herein will become apparent to those skilled in the art from the foregoing description and the accompanying figures. Such modifications are intended to fall within the scope of the appended claims.

What is claimed is:

1. A method for altering the body composition of a subject comprising administering a first composition comprising an effective amount of a GDF8 inhibitor and a second composition comprising an effective amount of an Activin A inhibitor to the subject.

2. The method of claim 1, wherein the altering of the body composition comprises inducing a reduction in fat mass in the subject comprising administering the first composition comprising an effective amount of a GDF8 inhibitor and the second composition comprising an effective amount of an Activin A inhibitor to the subject.

3. The method of claim 1 or claim 2, wherein the altering of the body composition comprises inducing an increase in muscle mass in the subject, the method comprising administering the first composition comprising an effective amount of the GDF8 inhibitor and the second composition comprising an effective amount of the Activin A inhibitor.

4. A method for altering the body composition of a subject comprising administering a composition comprising an effective amount of a GDF8 inhibitor and an effective amount of an Activin A inhibitor to the subject.

5. The method of claim 4, wherein the altering of the body composition comprises inducing a reduction in fat mass in a subject comprising administering the composition comprising an effective amount of the GDF8 inhibitor and an effective amount of the Activin A inhibitor to the subject.

6. The method of claim 3 or claim 4, wherein the altering of the body composition comprises -inducing an increase in muscle mass in the subject, the method comprising administering to the subject in need thereof the composition comprising an effective amount of the GDF8 inhibitor and an effective amount of the Activin A inhibitor.

7. The method of any one of claims 1-6, wherein the effective amount of a GDF8 inhibitor comprises a dosing regimen selected from the group consisting of at least 0.01 mg/kg to about 10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100 mg/kg.

8. The method of any one of claims 1-7, wherein the effective amount of an Activin A inhibitor comprises a dosing regimen selected from the group consisting of at least 0.01 mg/kg to about
10 gm/kg, 1 mg/kg to about 1 gm/kg, and 10 mg/kg to 100mg/kg.

9. The method of any one of claims 1-6, wherein the effective amount of a GDF8 inhibitor comprises a dosing regimen selected from a group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1 to about 5 mg/kg body weight.

10. The method of any one claims 1-6 or 9, wherein the effective amount of an Activin A inhibitor comprises a dosing regimen selected from a group consisting of a single dose of about 0.01 to about 20 mg/kg body weight, about 0.1 to about 10 mg/kg body weight, and about 0.1 to about 5 mg/kg body weight.

11. The method of any one of claims 7-10, wherein the effective amount of the GDF8 inhibitor is 6 mg/kg body weight of the subject.

12. The method of any one of claims 7-11, wherein the effective amount of the Activin A inhibitor is 3 mg/kg or 10 mg/kg body weight of the subject.

13. The method of any one of claims 1-3, wherein the first composition is formulated for intravenous, subcutaneous, or oral administration.

14. The method of any one of claims 1-3, wherein the second composition is formulated for intravenous, subcutaneous, or oral administration.

15. The method of any one of claims 1-3, wherein the first and second compositions are combined into a third composition prior to administration.

16. The method of claim 15, wherein the third composition is formulated for intravenous, subcutaneous, or oral administration.

17. The method of any one of claims 3-6, wherein the composition is formulated for intravenous, subcutaneous, or oral administration.

18. The method of any one of claims 1-17, further comprising measuring total fat mass and/or android fat mass in the subject before administration.

19. The method of claim 18, further comprising measuring total fat mass and /or android fat mass in the subject after administration, and administering the composition until the subject has a reduction in total fat mass and/or android fat mass of at least about 2% to 8%, at least about 2.5% to 6%, at least about 3% to 4%, or at least about 3.5%.

20. The method of any one of claims 1-19, wherein the GDF8 inhibitor is an isolated antibody or an antigen-binding fragment thereof that specifically binds to GDF8.

21. The method of claim 20, wherein the antibody or antigen-binding fragment that specifically binds GDF8 comprises the heavy chain complementarity determining regions (HCDRs) of a heavy chain variable region (HCVR) comprising SEQ ID NO:360, and the light chain complementarity determining regions (LCDRs) of a light chain variable region (LCVR) comprising SEQ ID NO:368.

22. The method of claim 20 or 21, wherein the antibody or antigen-binding fragment that specifically binds GDF8 comprises three HCDRs comprising SEQ ID NO:362, SEQ ID

NO:364, and SEQ ID NO:366, and three LCDRs comprising SEQ ID NO:370, SEQ ID NO:372, and SEQ ID NO:374.

23. The method of any one of claims 1-22, wherein the Activin A inhibitor is an isolated antibody or antigen-binding fragment thereof that specifically binds Activin A.

24. The method of claim 23, wherein the antibody or antigen-binding fragment that specifically binds Activin A comprises the heavy chain complementarity determining regions (HCDRs) of a heavy chain variable region (HCVR) comprising SEQ ID NO:553, and the light chain complementarity determining regions (LCDRs) of a light chain variable region (LCVR) comprising SEQ ID NO:537.

25. The method of claim 23 or 24, wherein the antibody or antigen-binding fragment that specifically binds Activin A comprises three HCDRs comprising SEQ ID NO:555, SEQ ID NO:557, and SEQ ID NO:559, and three LCDRs comprising SEQ ID NO:539, SEQ ID NO:541, and SEQ ID NO:543.

26. The method of any one of claims 1-25, wherein the Activin A inhibitor is in an amount selected from the group consisting of between 100% to 200% of the amount of the GDF8 inhibitor, between 100% and 250% of the amount of the GDF8 inhibitor, between 100% and 300% of the amount of the GDF8 inhibitor, and between 100% and 400% by weight of the amount of the GDF8 inhibitor.

27. The method of claim 26, wherein the amount of the Activin A inhibitor is about 1.5 to 2.0 times as large by weight as the amount of the GDF8 inhibitor.

28. The method of any one of claims 1-27, wherein a reduction of fat mass in the subject is a reduction in total fat mass as measured by DXA (Dual-energy X-ray absorptiometry).

29. The method of any one of claims 1-28, wherein a reduction of fat mass in the subject is a reduction in android fat mass as measured by DXA (Dual-energy X-ray absorptiometry).

30. The method of any one of claims 1-29, wherein the subject experiences an increase in muscle volume after administration.

31. The method of any one of claims 1-30, wherein the subject does not have a muscle wasting condition or disease.

32. A GDF8 inhibitor for use in a method for treating a disease or disorder characterized by increased fat mass, wherein the method comprises administrating to a subject the GDF8 inhibitor and an Activin A inhibitor.

33. An Activin A inhibitor for use in a method for treating a disease or disorder characterized by increased fat mass, wherein the method comprises administrating to a subject the Activin A inhibitor and a GDF8 inhibitor.

34. A non-therapeutic method for decreasing fat mass in a subject, the method comprising administering to the subject an Activin A inhibitor and a GDF8 inhibitor.

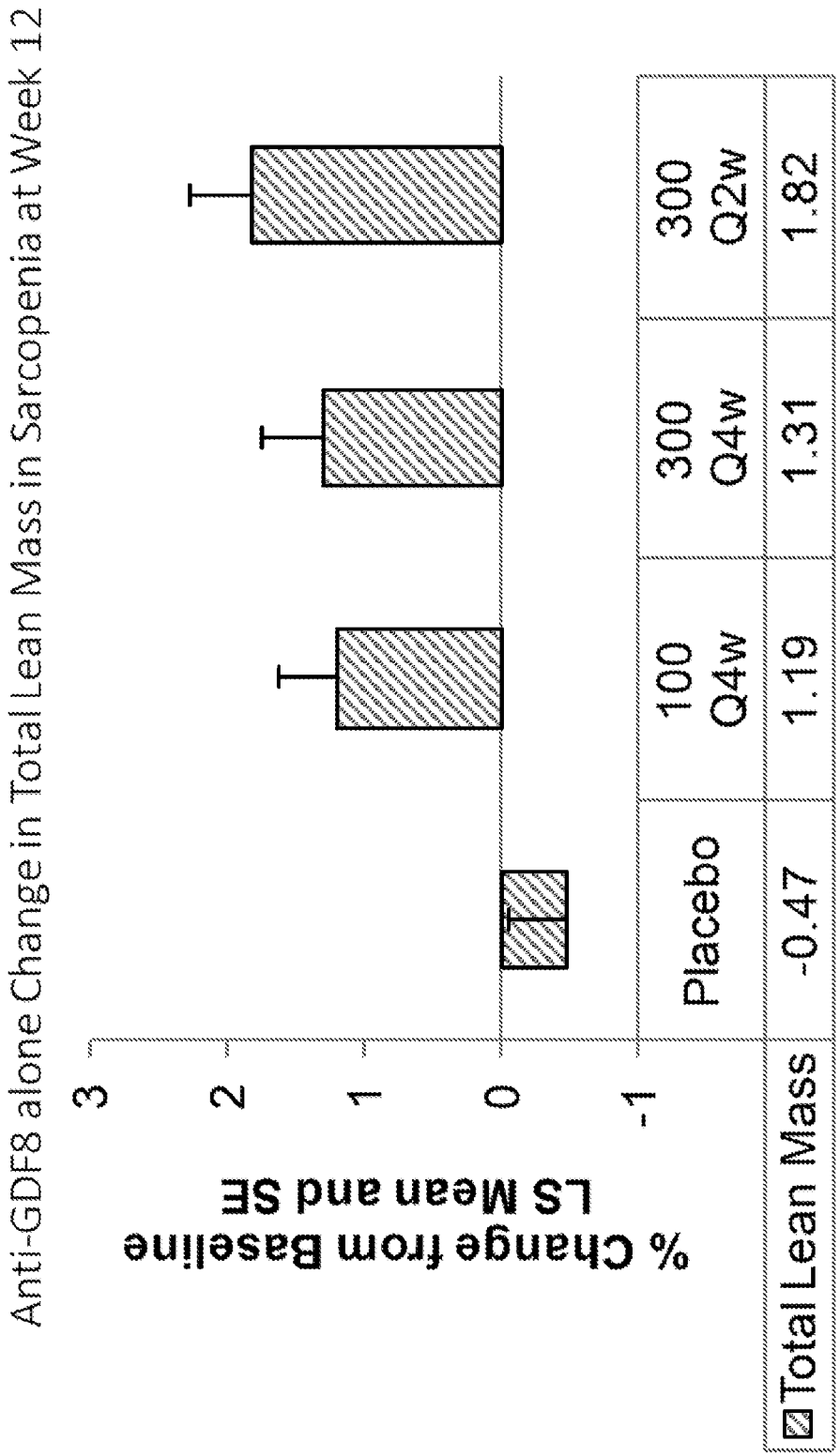

Anti-GDF8 alone Change in Total Lean Mass in Sarcopenia at Week 12
% Change from Baseline LS Mean and SE
3
2
1
0
-1
Placebo
100 Q4w
300 Q4w
300 Q2w
Total Lean Mass
-0.47
1.19
1.31
1.82

Figure 1

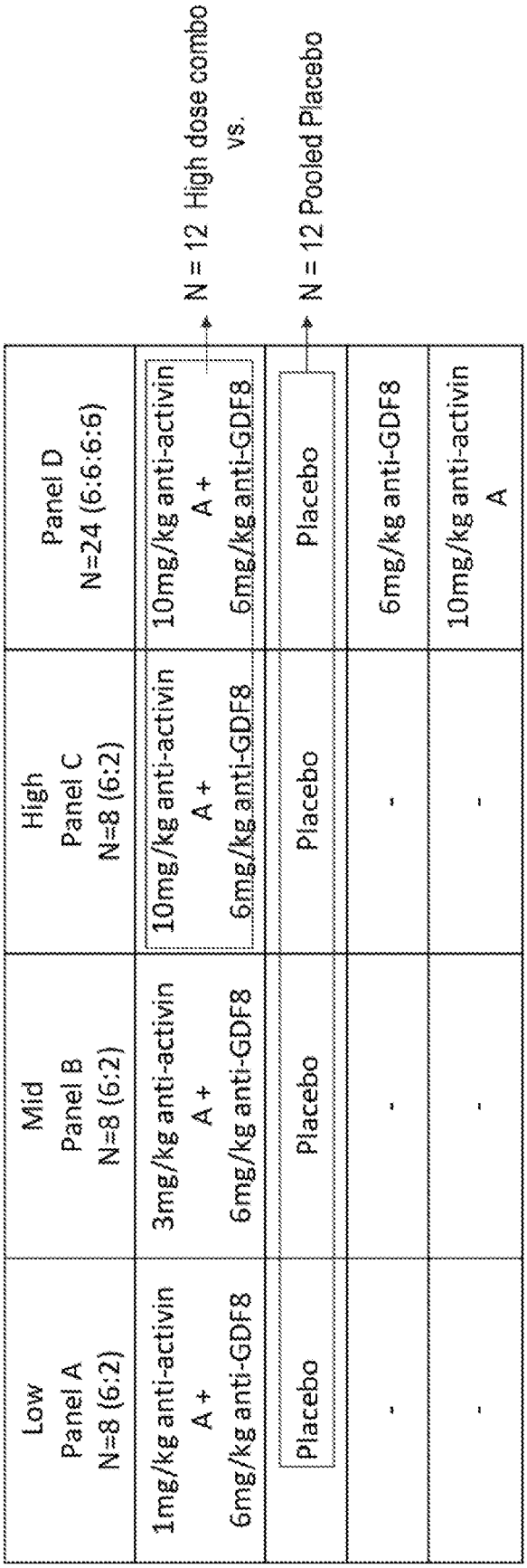

Low
Panel A
N=8 (6:2)
Mid
Panel B
N=8 (6:2)
High
Panel C
N=8 (6:2)
Panel D
N=24 (6:6:6:6)
1mg/kg anti-activin A + 6mg/kg anti-GDF8
3mg/kg anti-activin A + 6mg/kg anti-GDF8
10mg/kg anti-activin A + 6mg/kg anti-GDF8
10mg/kg anti-activin A + 6mg/kg anti-GDF8
Placebo
Placebo
Placebo
Placebo
-
-
-
6mg/kg anti-GDF8
-
-
-
10mg/kg anti-activin A
N = 12 High dose combo vs.
N = 12 Pooled Placebo

Figure 2A

Figure 2C

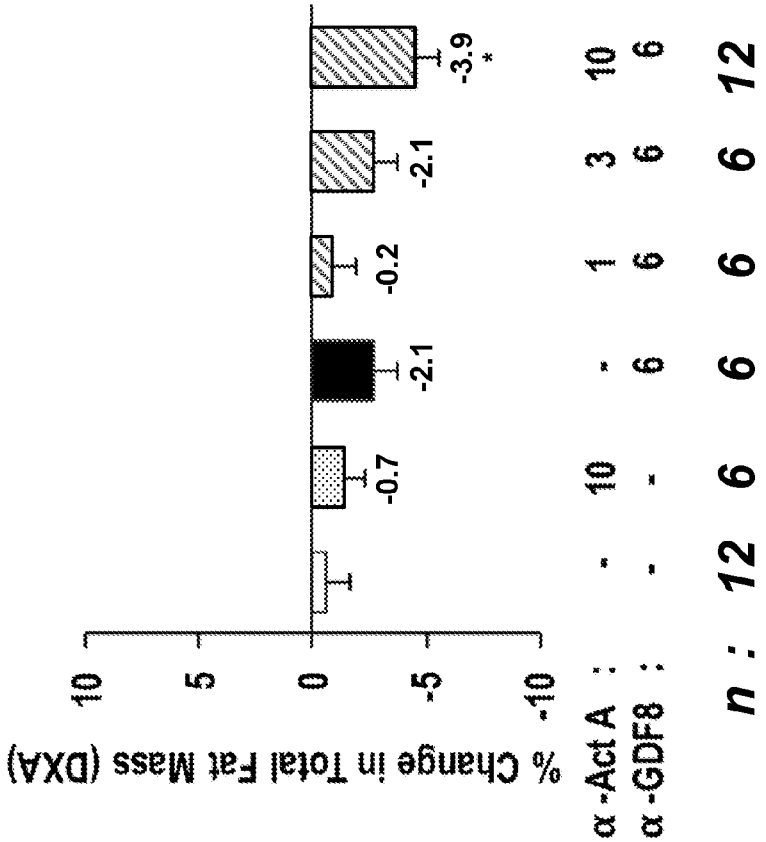
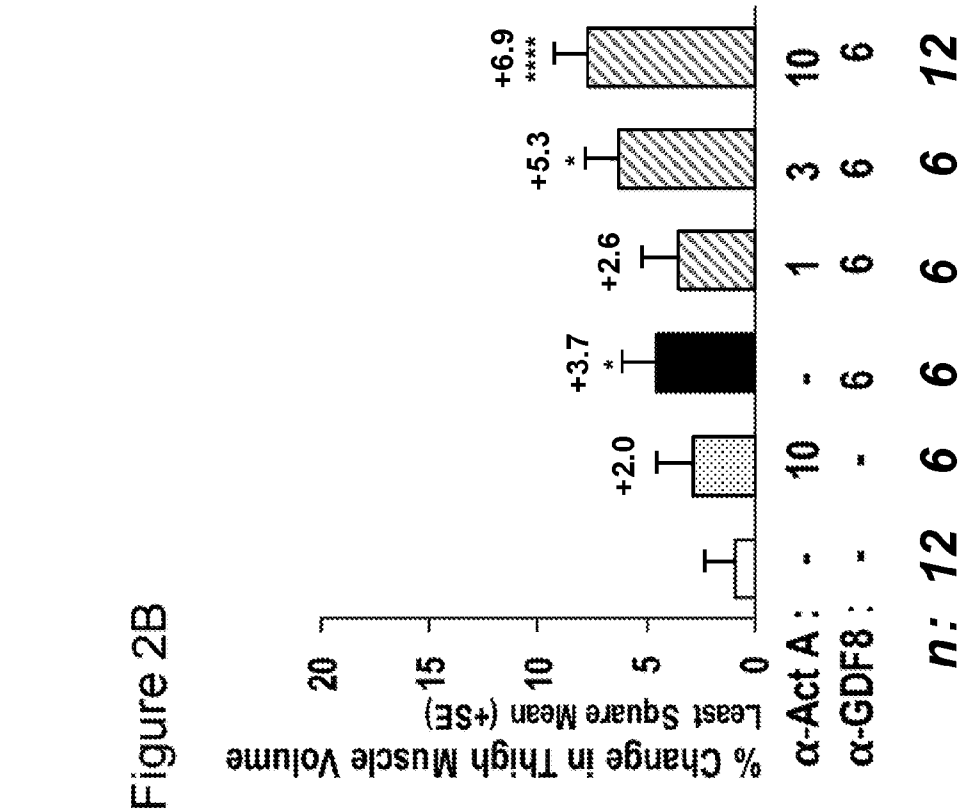


Figure 2B



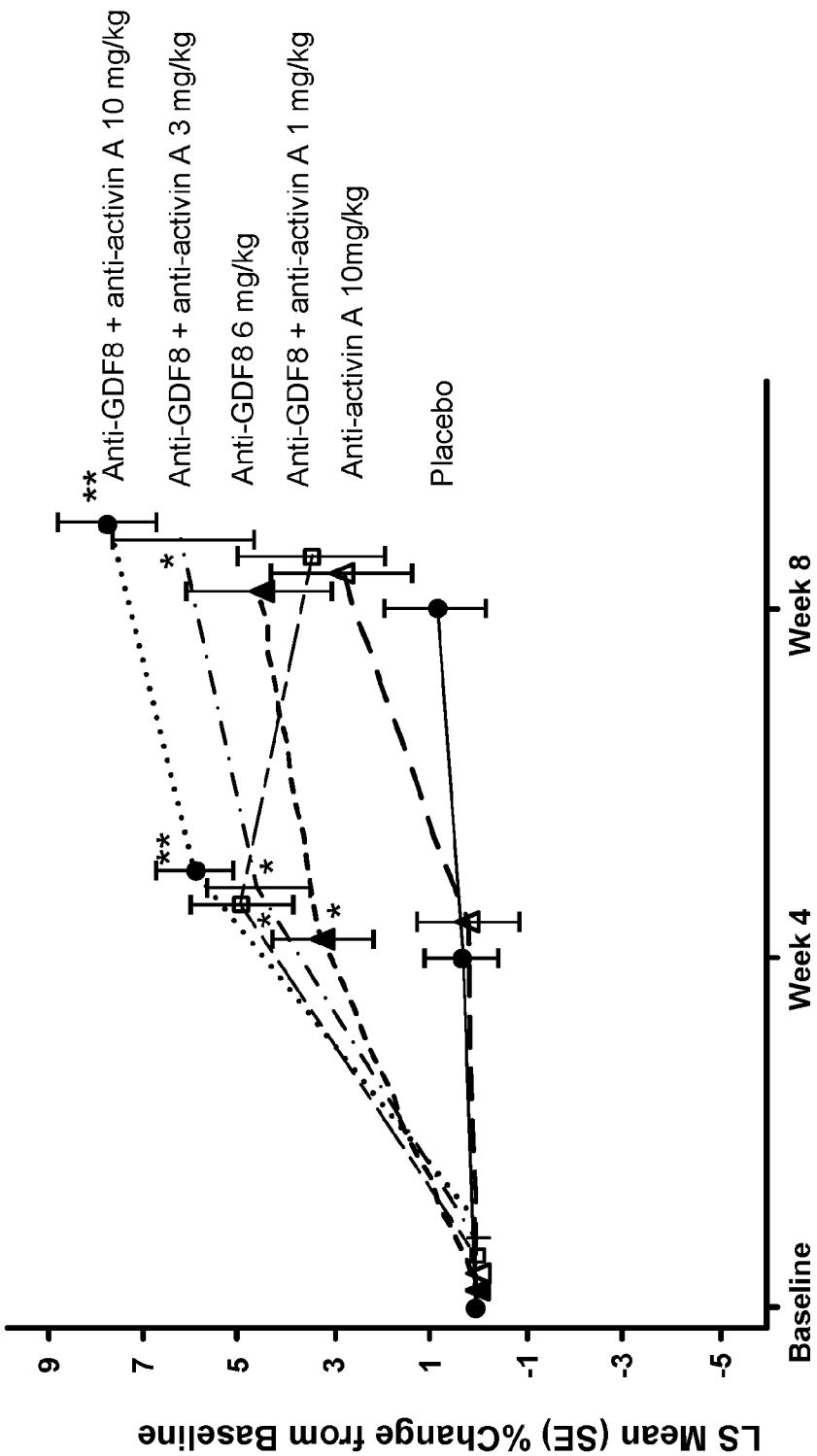

Anti-GDF8 + anti-activin A 10 mg/kg
Anti-GDF8 + anti-activin A 3 mg/kg
Anti-GDF8 6 mg/kg
Anti-GDF8 + anti-activin A 1 mg/kg
Anti-activin A 10mg/kg
Placebo
LS Mean (SE) %Change from Baseline
9
7
5
3
1
-1
-3
-5
Baseline
Week 4
Week 8

Figure 3

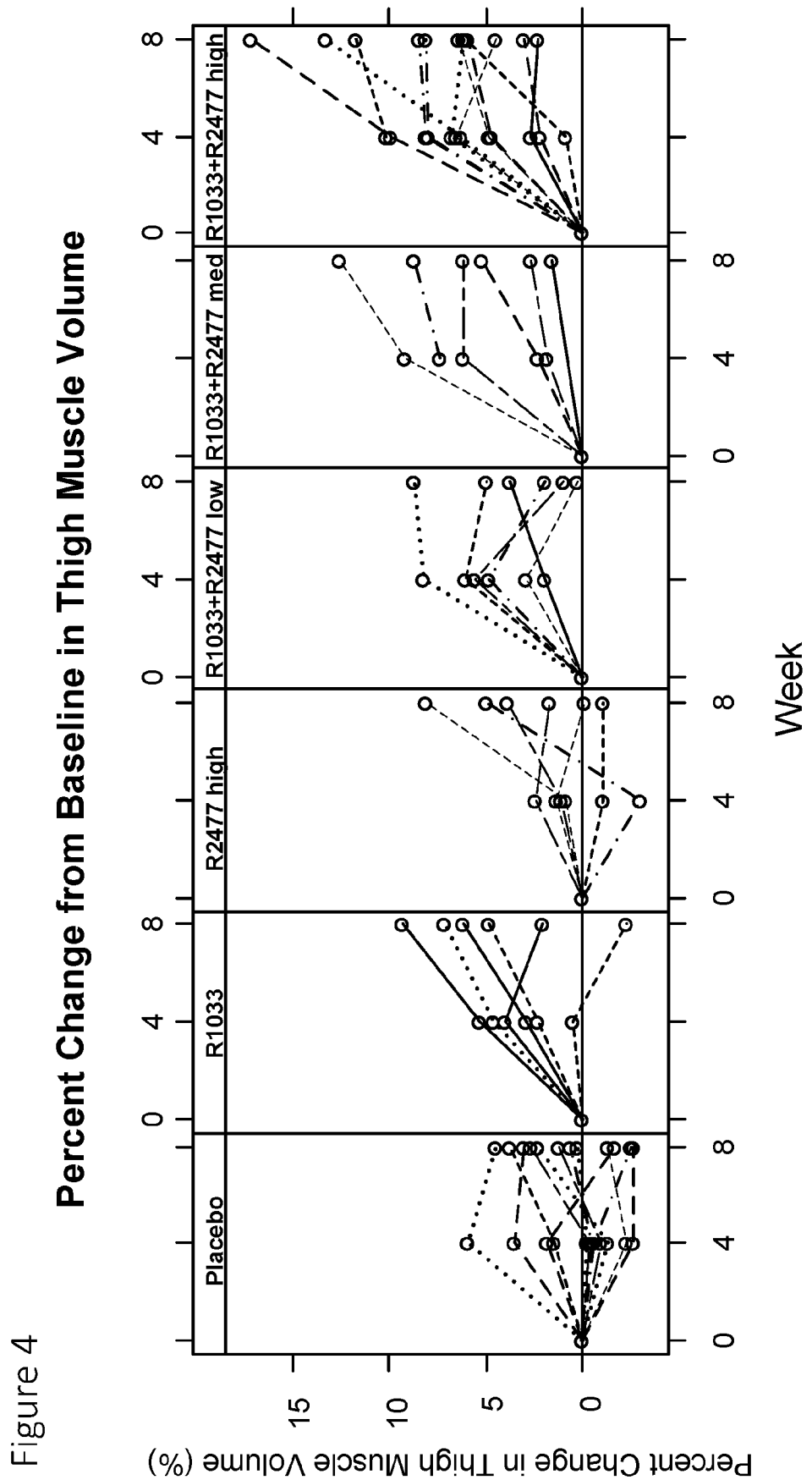

Percent Change from Baseline in Thigh Muscle Volume
Placebo
R1033
R2477 high
R1033+R2477 low
R1033+R2477 med
R1033+R2477 high
Percent Change in Thigh Muscle Volume (%)
Week
0
5
10
15
0
4
8

Figure 4

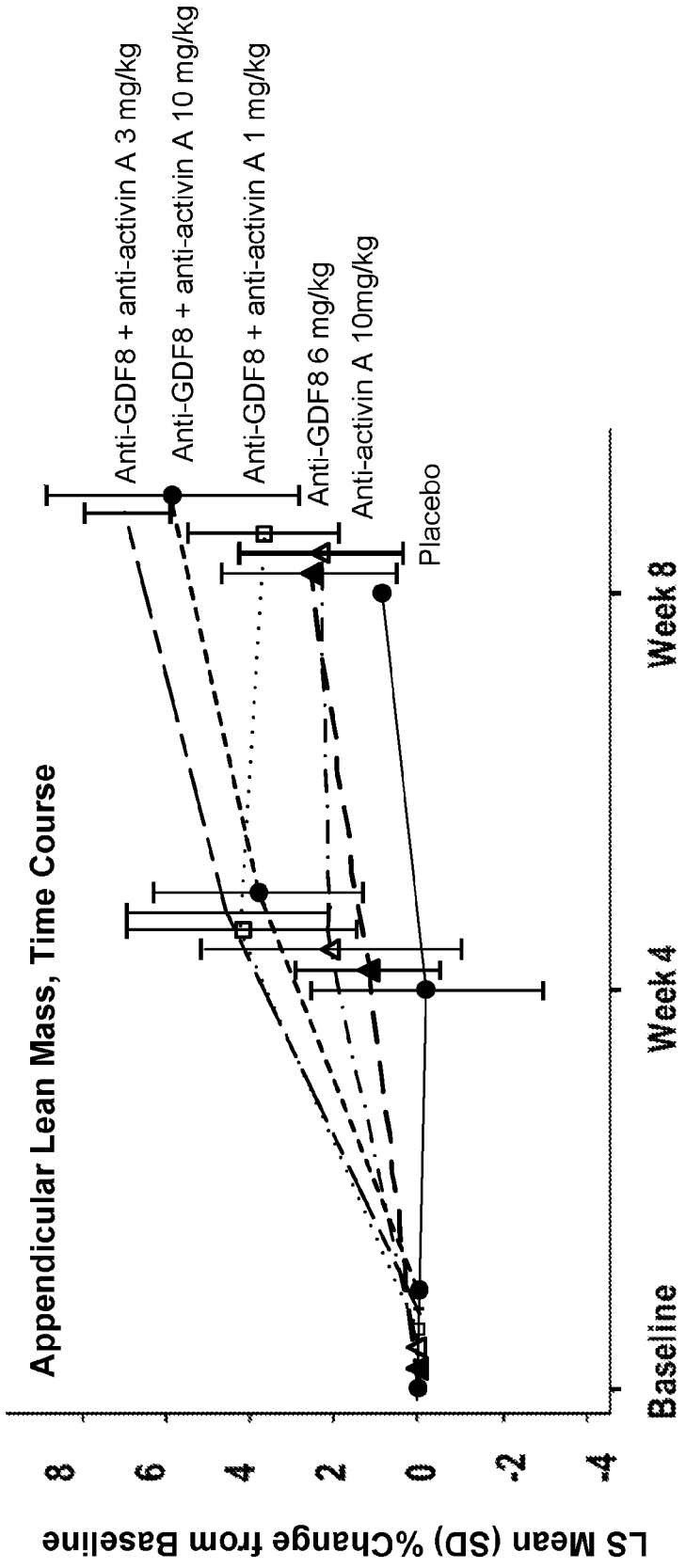

Appendicular Lean Mass, Time Course
LS Mean (SD) %Change from Baseline
8
6
4
2
0
-2
-4
Baseline
Week 4
Week 8
Anti-GDF8 + anti-activin A 3 mg/kg
Anti-GDF8 + anti-activin A 10 mg/kg
Anti-GDF8 + anti-activin A 1 mg/kg
Anti-GDF8 6 mg/kg
Anti-activin A 10mg/kg
Placebo

Figure 5

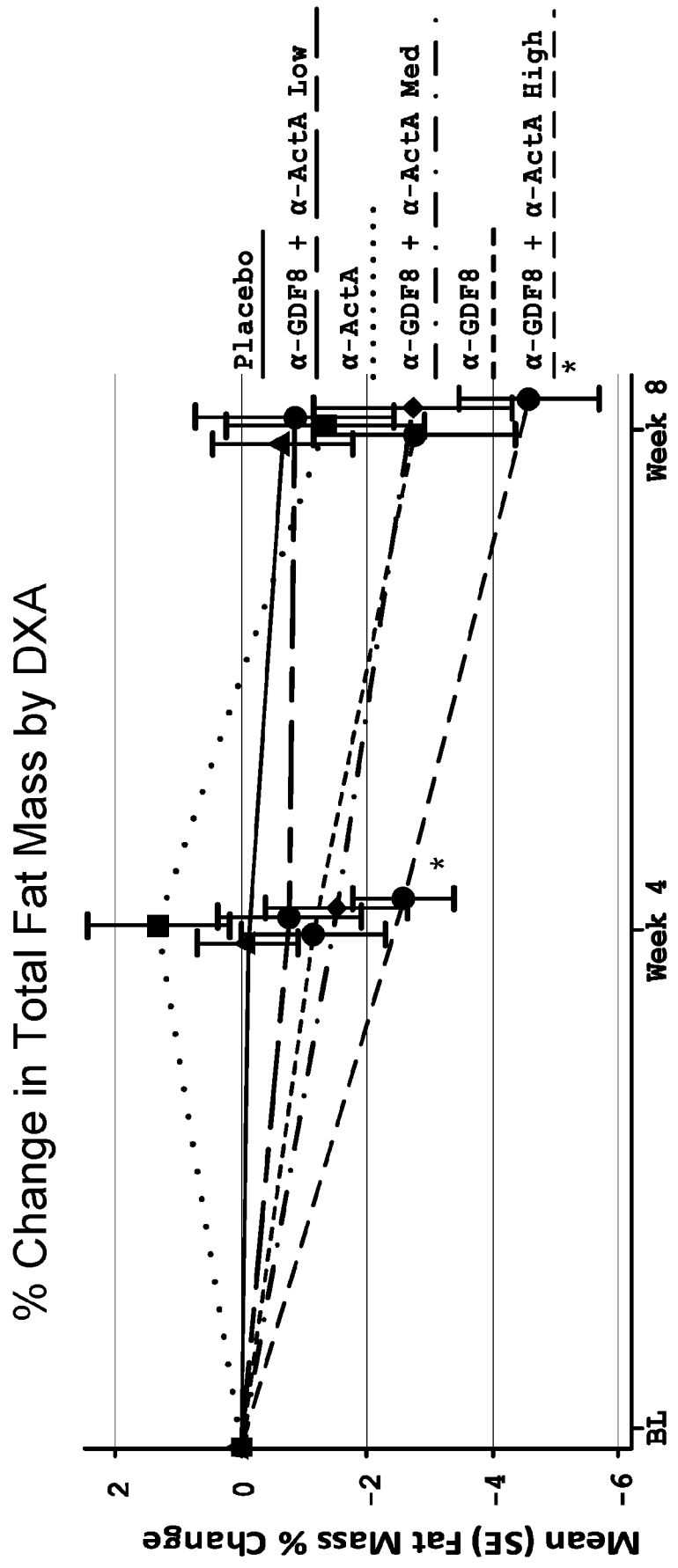

% Change in Total Fat Mass by DXA
Placebo
α-GDF8 + α-ActA Low
α-ActA
α-GDF8 + α-ActA Med
α-GDF8
α-GDF8 + α-ActA High
*
*
Mean (SE) Fat Mass % Change
2
0
-2
-4
-6
BL
Week 4
Week 8
Study Week

Figure 6

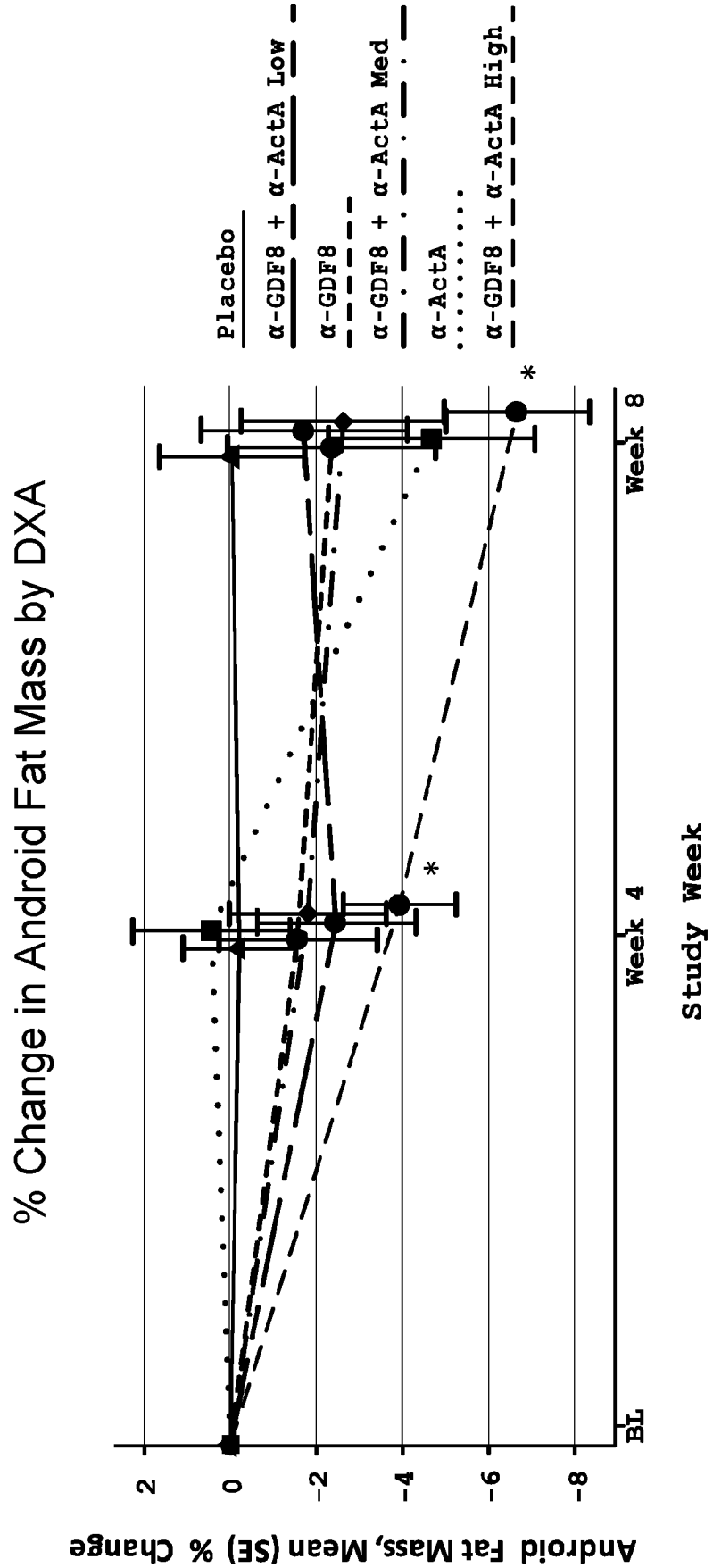

% Change in Android Fat Mass by DXA
Placebo
α-GDF8 + α-ActA Low
α-GDF8
α-GDF8 + α-ActA Med
α-ActA
α-GDF8 + α-ActA High
Android Fat Mass, Mean (SE) % Change
2
0
-2
-4
-6
-8
BL
Week 4
Week 8
Study Week
*
*

Figure 7

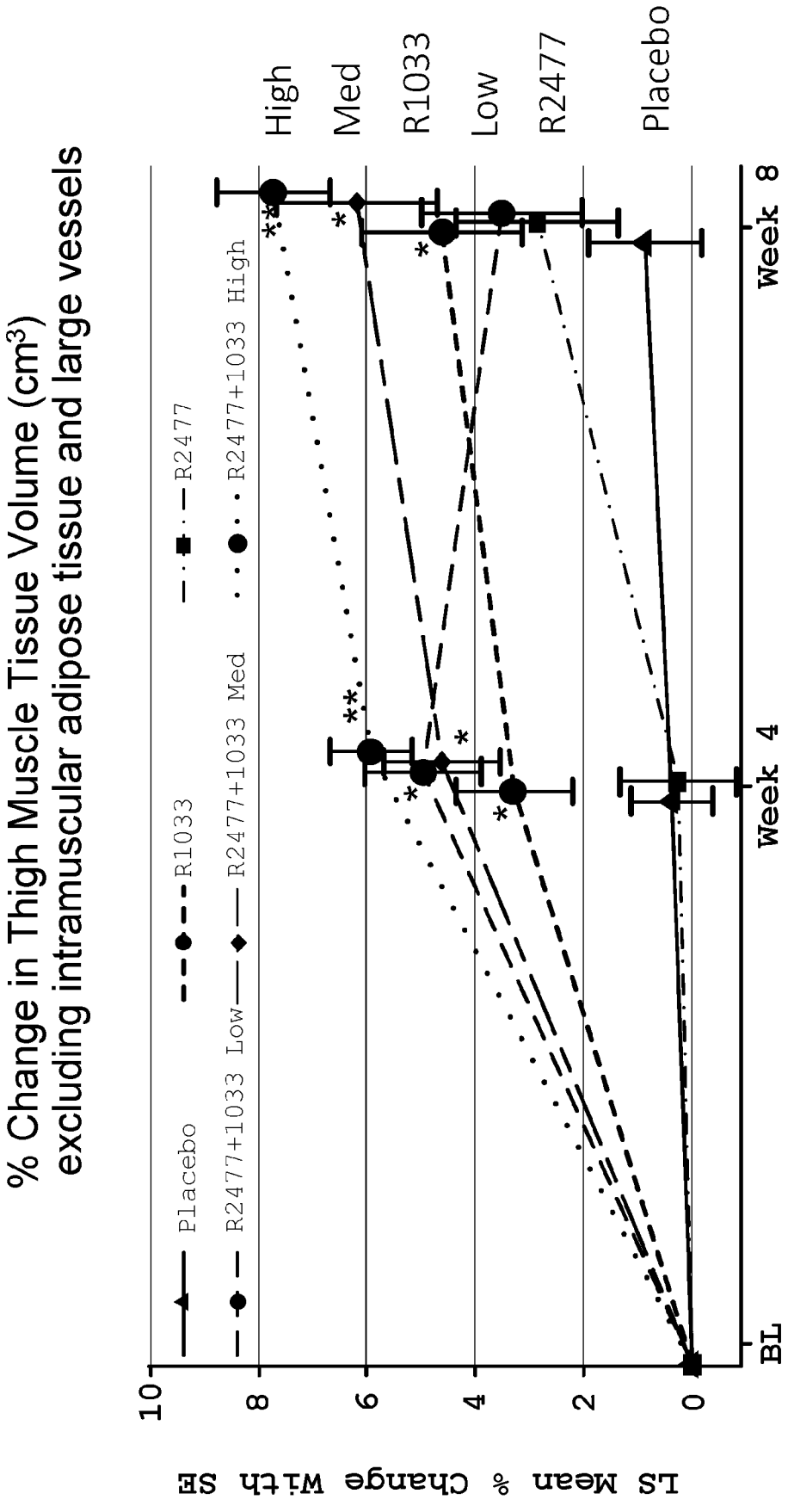

% Change in Thigh Muscle Tissue Volume (cm3)
excluding intramuscular adipose tissue and large vessels
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
High
Med
R1033
Low
R2477
Placebo
LS Mean % Change With SE
10
8
6
4
2
0
BL
Week 4
Week 8
Study Week

Figure 8

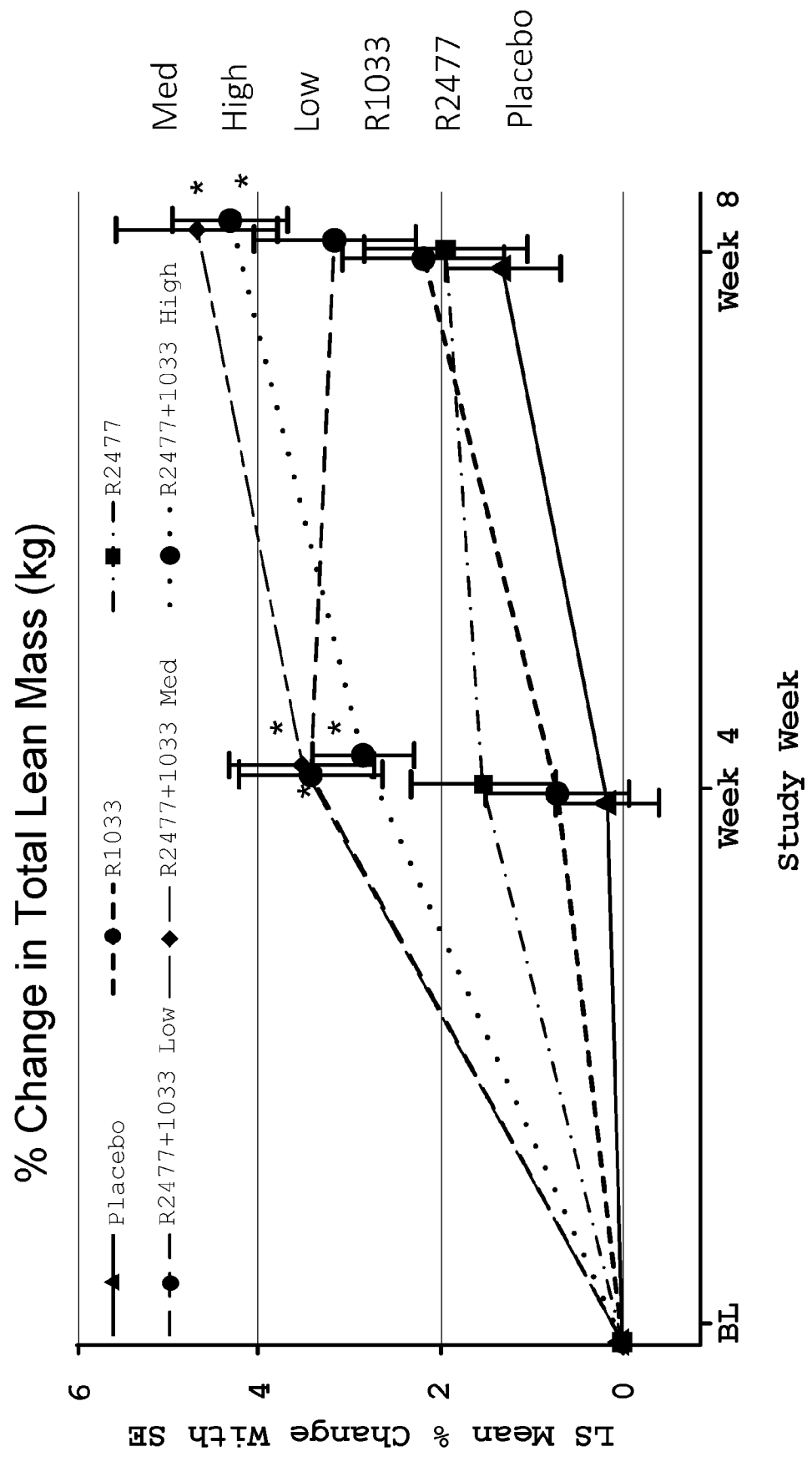

% Change in Total Lean Mass (kg)
LS Mean % Change with SE
Study Week
BL
Week 4
Week 8
0
2
4
6
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
Placebo
R1033
R2477
Low
High
Med
*

Figure 9

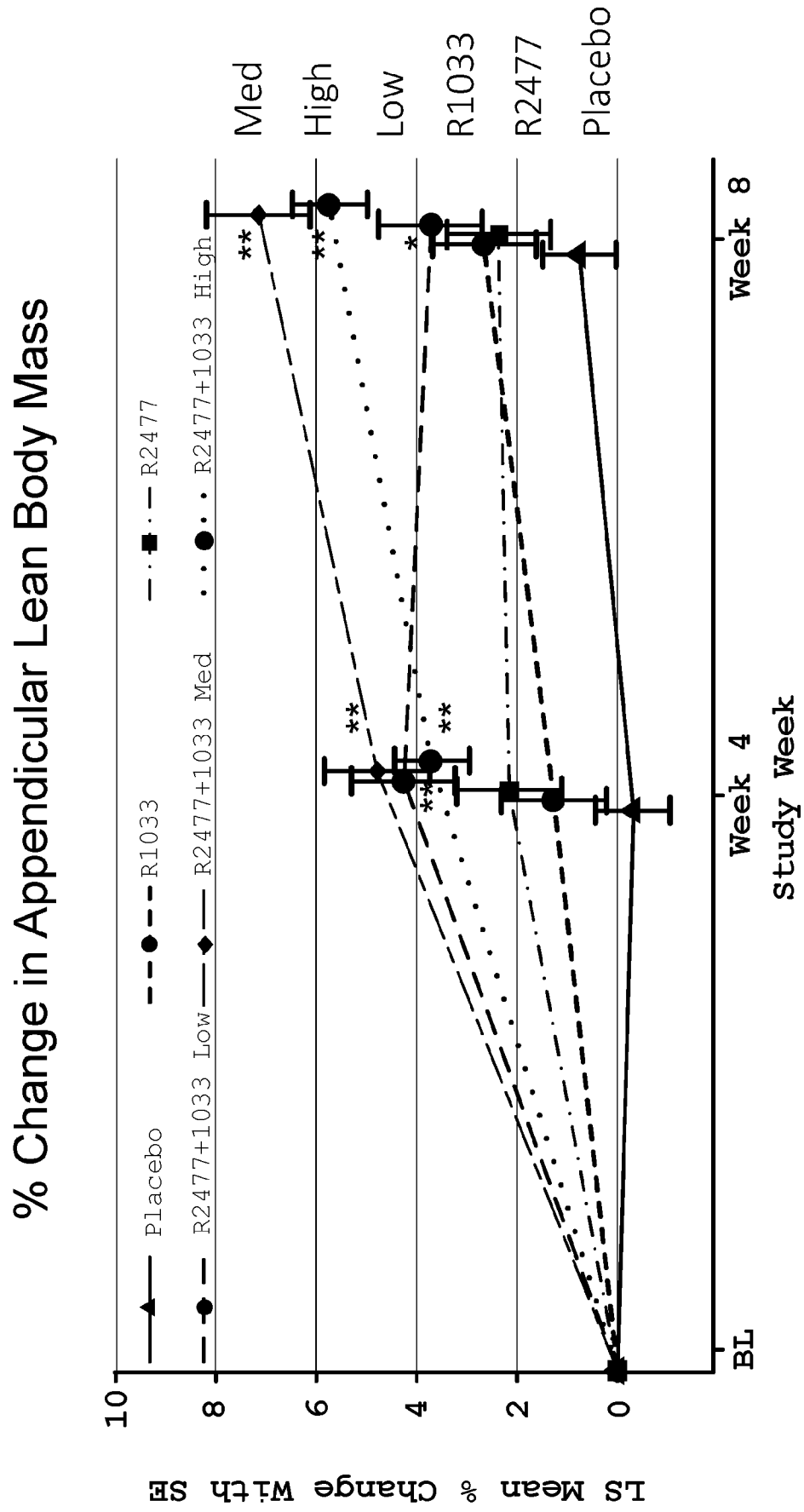

% Change in Appendicular Lean Body Mass
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
LS Mean % Change With SE
10
8
6
4
2
0
BL
Week 4
Week 8
Study Week
Med
High
Low
R1033
R2477
Placebo
**
**
**
**
**
*

Figure 10

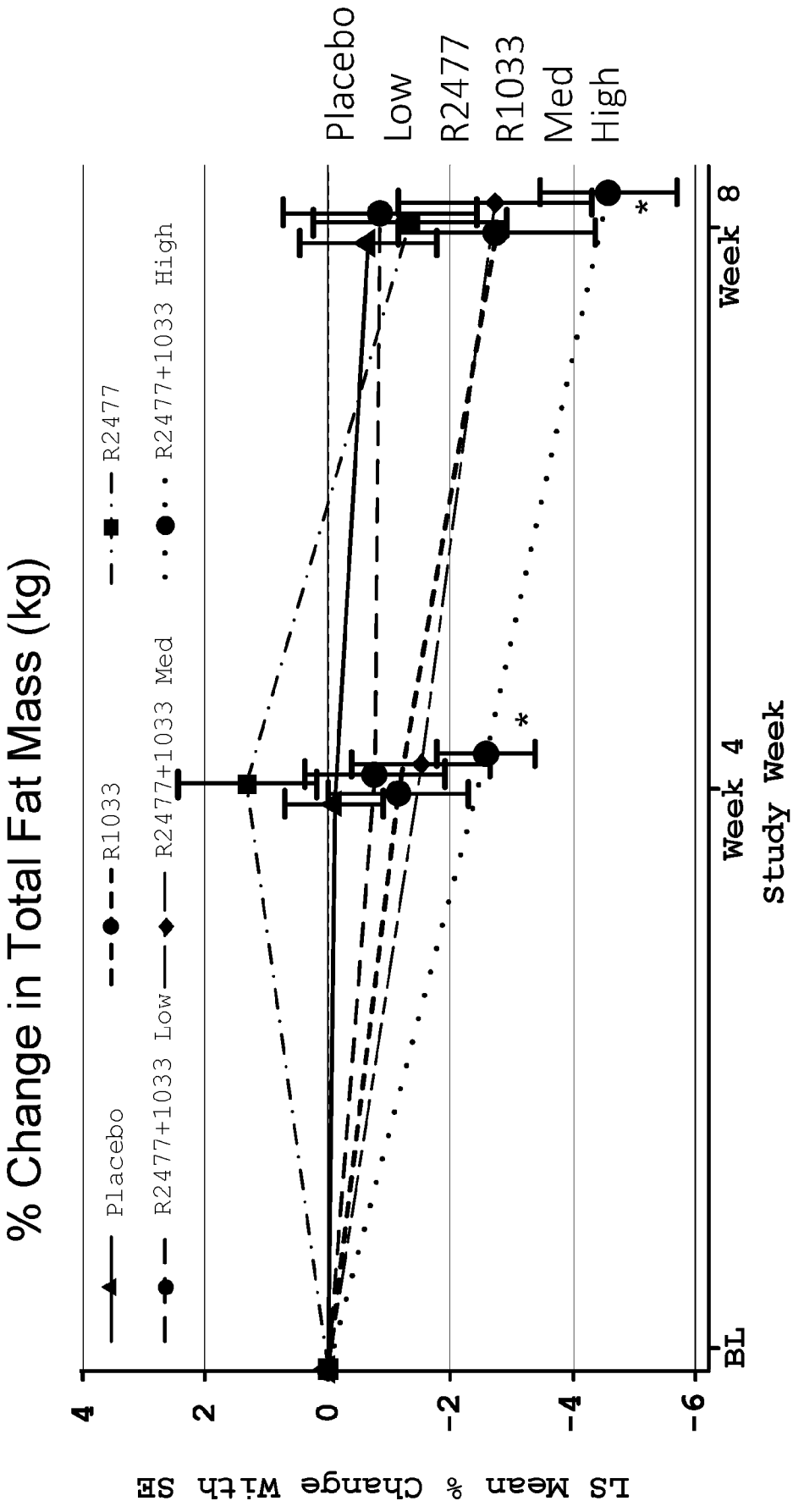

% Change in Total Fat Mass (kg)
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
Placebo
Low
R2477
R1033
Med
High
LS Mean % Change with SE
4
2
0
-2
-4
-6
BL
Week 4
Week 8
Study Week
*
*

Figure 11

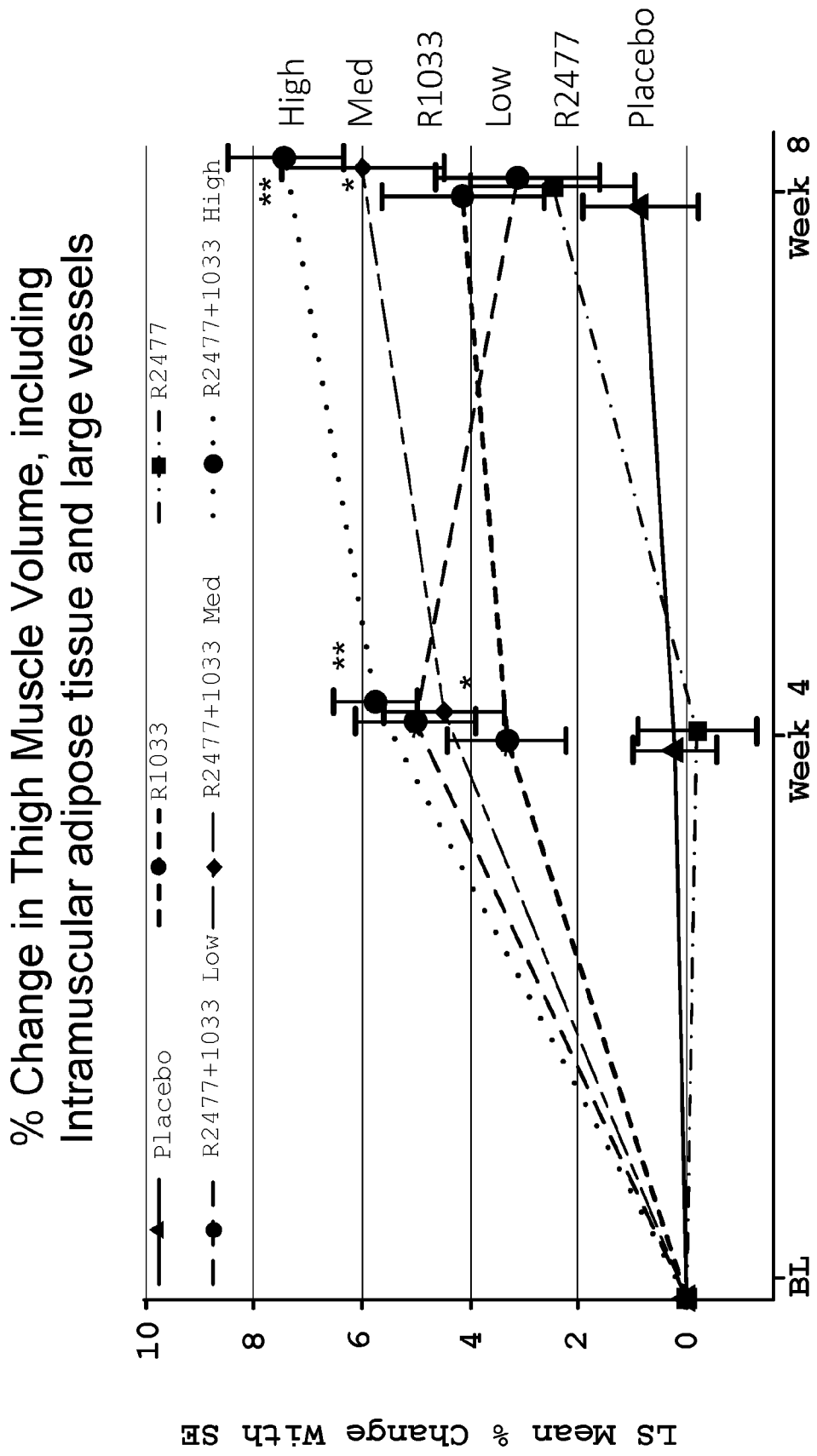

% Change in Thigh Muscle Volume, including Intramuscular adipose tissue and large vessels
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
LS Mean % Change With SE
10
8
6
4
2
0
BL
Week 4
Week 8
High
Med
R1033
Low
R2477
Placebo
**
*
**
*

Figure 12

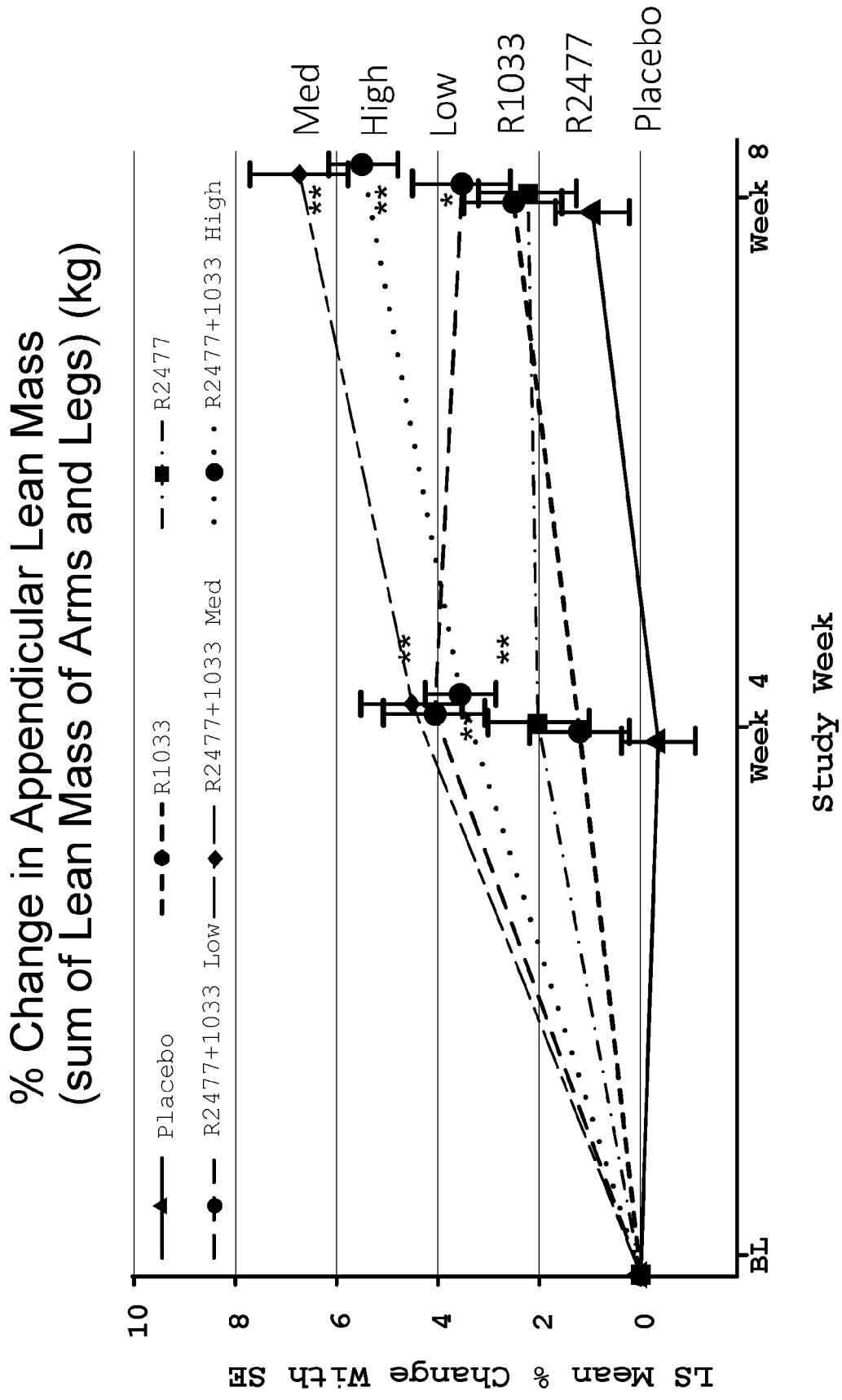

% Change in Appendicular Lean Mass
(sum of Lean Mass of Arms and Legs) (kg)
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
LS Mean % Change with SE
10
8
6
4
2
0
BL
Week 4
Week 8
Study Week
Med
High
Low
R1033
R2477
Placebo
*
**

Figure 13

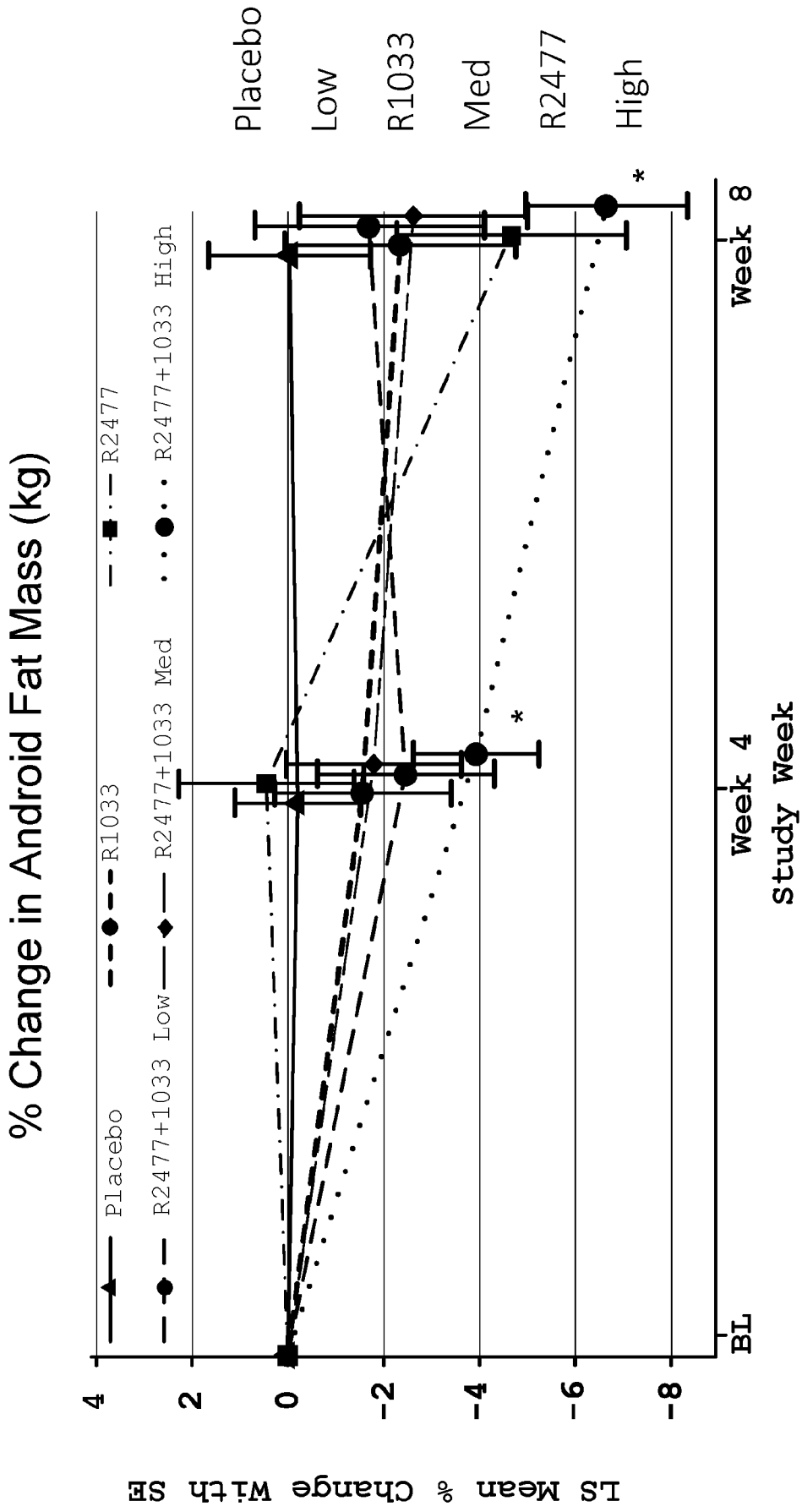

% Change in Android Fat Mass (kg)
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
Placebo
Low
R1033
Med
R2477
High
LS Mean % Change with SE
4
2
0
-2
-4
-6
-8
BL
Week 4
Week 8
Study Week
*
*

Figure 14

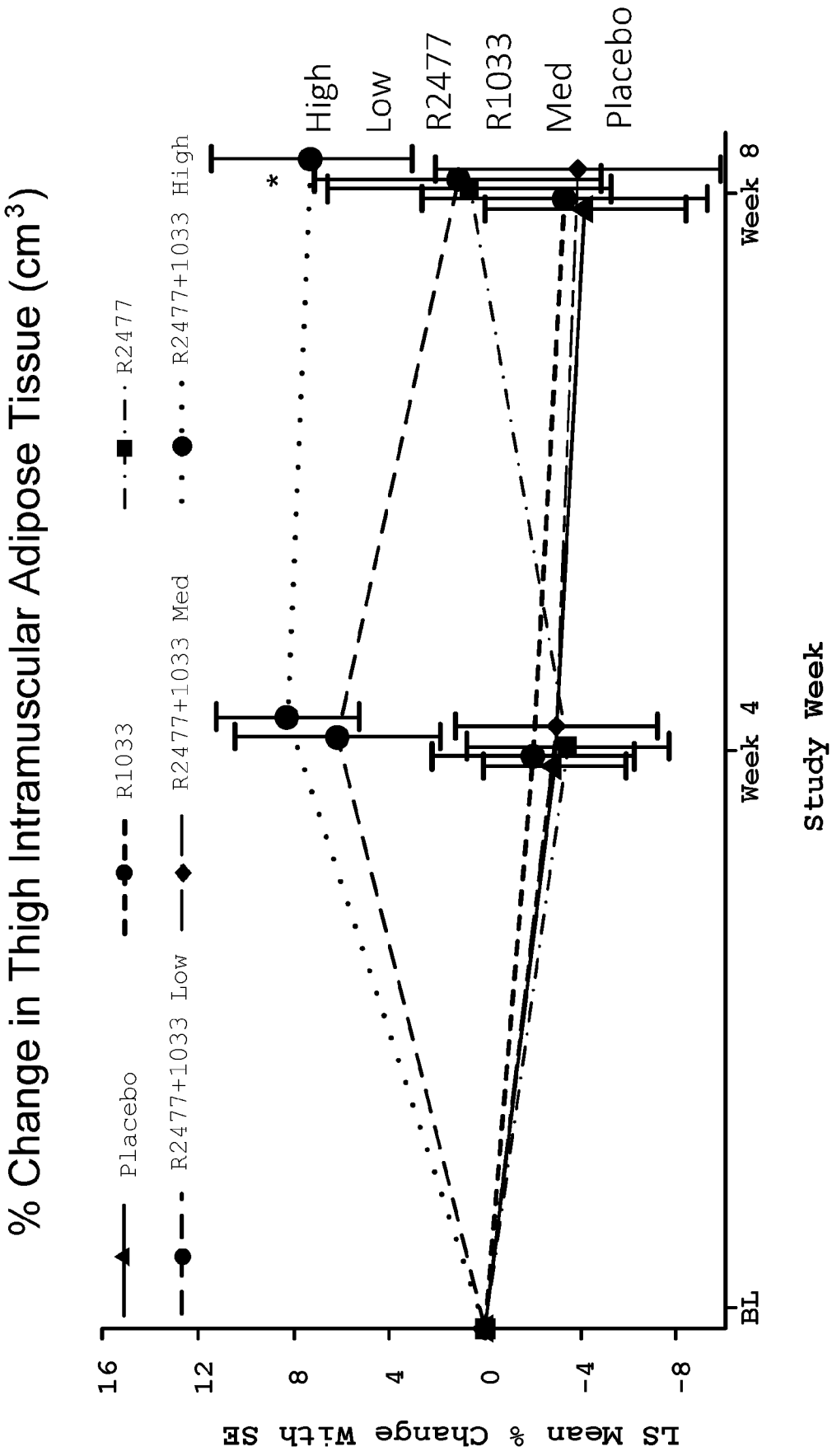

% Change in Thigh Intramuscular Adipose Tissue (cm3)
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
LS Mean % Change with SE
16
12
8
4
0
-4
-8
BL
Week 4
Week 8
Study Week
*
High
Low
R2477
R1033
Med
Placebo

Figure 15

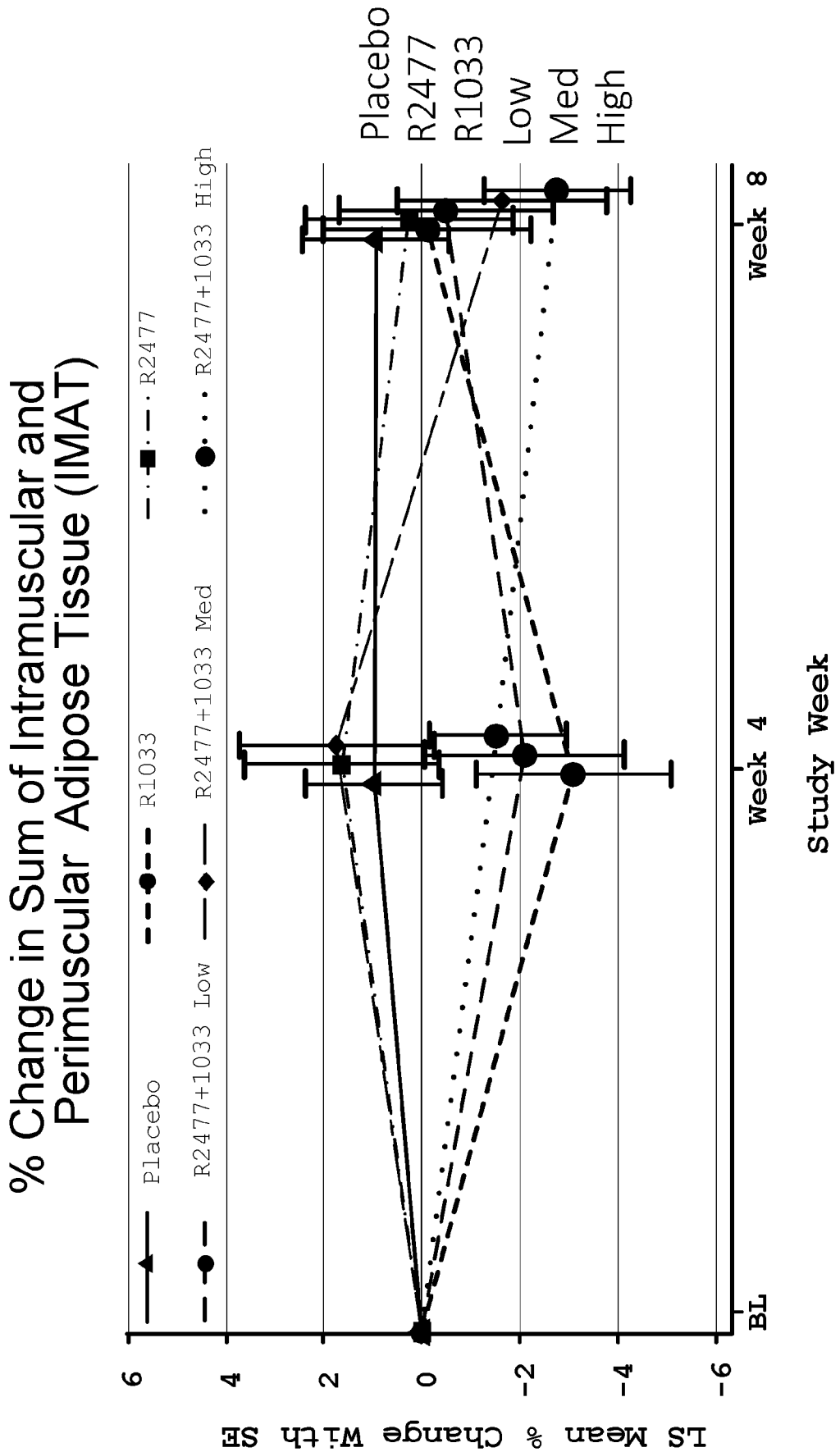

% Change in Sum of Intramuscular and Perimuscular Adipose Tissue (IMAT)
Placebo
R2477
R1033
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
Placebo
R2477
R1033
Low
Med
High
LS Mean % Change with SE
6
4
2
0
-2
-4
-6
BL
Week 4
Week 8
Study Week

Figure 16

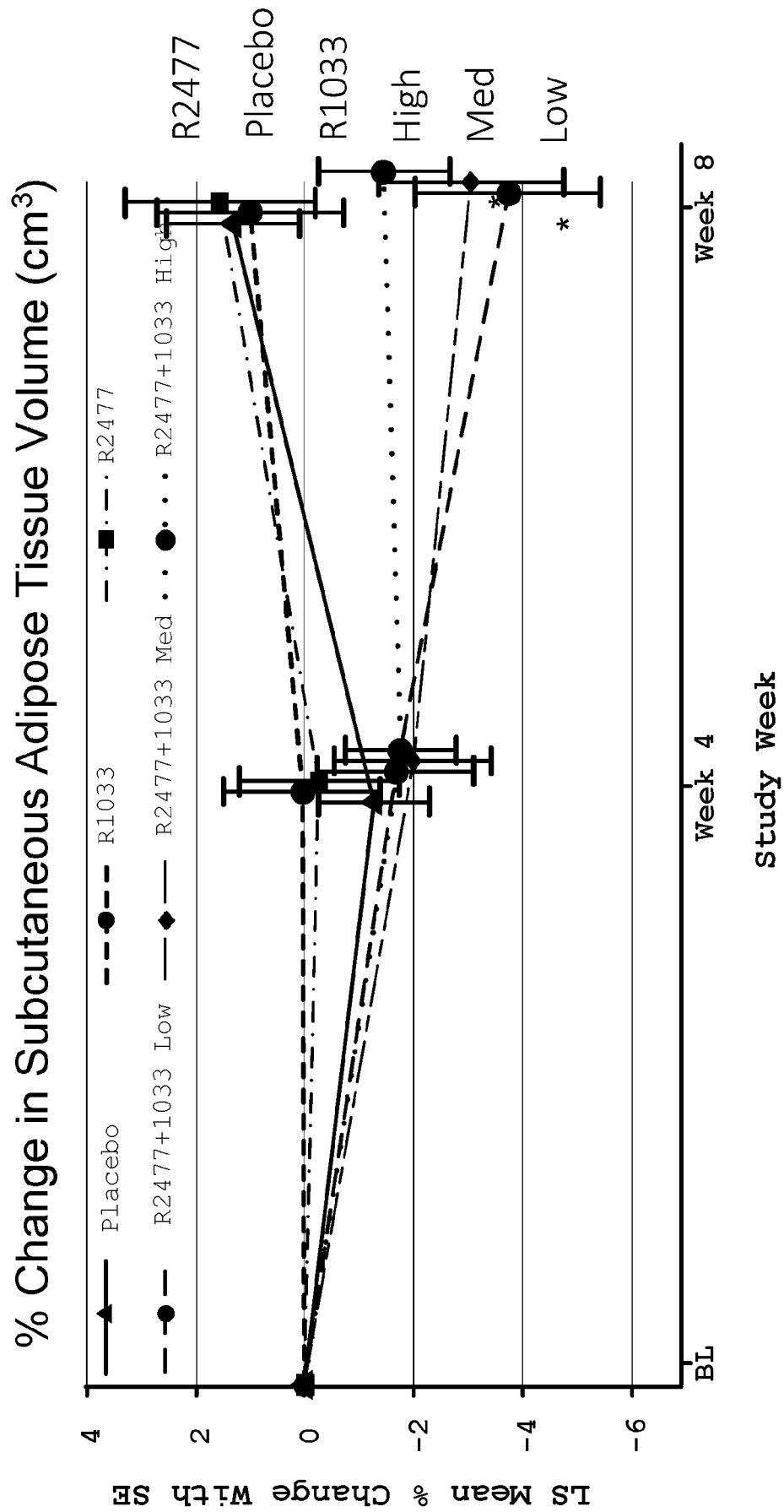

% Change in Subcutaneous Adipose Tissue Volume (cm3)
Placebo
R1033
R2477
R2477+1033 Low
R2477+1033 Med
R2477+1033 High
LS Mean % Change with SE
4
2
0
-2
-4
-6
BL
Week 4
Week 8
Study Week
R2477
Placebo
R1033
High
Med
Low
*
*

Figure 17

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2019/020330

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61P21/00 A61K39/395
ADD.

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

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61P C07K A61K

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, BIOSIS, EMBASE, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Ester Latres: "Activin A more prominently regulates muscle mass in primates than does GDF8", , 28 July 2017 (2017-07-28), pages 1-13, XP055584641, Retrieved from the Internet: URL:https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5414365/ [retrieved on 2019-04-30]	1-10, 13-25, 28-34
Y	page 3 - page 4; figures 1,6 page 10, paragraph 3	1-34

X	US 2013/122007 A1 (STITT TREVOR [US] ET AL) 16 May 2013 (2013-05-16)	1-10, 12-23, 28-34
Y	paragraphs [0009], [0043], [0070], [0071], [0072]; sequence 9	1-34

	-/--	

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
2 May 2019	16/05/2019

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Zellner, Eveline

1

Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2019/020330

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X Y	US 2010/183624 A1 (SEEHRA JASBIR [US] ET AL) 22 July 2010 (2010-07-22) paragraphs [0012], [13142], [0207] -----	1-6, 28-34 1-34
X Y	US 2015/037339 A1 (GROMADA JESPER [US] ET AL) 5 February 2015 (2015-02-05) paragraph [0110] - paragraph [0115] paragraph [0122]; claim 41 -----	1-25, 28-34 1-34
Y	WO 2016/168613 A1 (REGENERON PHARMA [US]) 20 October 2016 (2016-10-20) paragraphs [0006], [0007], [0008] -----	1-34

1

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2019/020330

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2013122007 A1	16-05-2013	AU 2012339722 A1	05-06-2014
		CA 2855822 A1	23-05-2013
		CY 1119994 T1	12-12-2018
		DK 2780368 T3	05-02-2018
		EP 2780368 A1	24-09-2014
		ES 2663946 T3	17-04-2018
		HK 1202559 A1	02-10-2015
		HR P20180201 T1	09-03-2018
		HU E038570 T2	29-10-2018
		JP 6177249 B2	09-08-2017
		JP 2014534239 A	18-12-2014
		KR 20140108527 A	11-09-2014
		LT 2780368 T	12-03-2018
		MX 350355 B	04-09-2017
		PL 2780368 T3	29-06-2018
		PT 2780368 T	22-03-2018
		SI 2780368 T1	30-04-2018
		US 2013122007 A1	16-05-2013
		US 2015010568 A1	08-01-2015
		US 2017008956 A1	12-01-2017
		US 2017226197 A1	10-08-2017
		US 2018171006 A1	21-06-2018
		WO 2013074557 A1	23-05-2013
US 2010183624 A1	22-07-2010	AU 2010204985 A1	04-08-2011
		AU 2016200913 A1	03-03-2016
		CA 2749544 A1	22-07-2010
		EP 2387412 A1	23-11-2011
		US 2010183624 A1	22-07-2010
		US 2012156204 A1	21-06-2012
		US 2015056200 A1	26-02-2015
		US 2016264681 A1	15-09-2016
		WO 2010083034 A1	22-07-2010
US 2015037339 A1	05-02-2015	AU 2014296243 A1	03-03-2016
		BR 112016002164 A2	12-09-2017
		CA 2920071 A1	05-02-2015
		CL 2016000251 A1	14-07-2017
		CN 105592858 A	18-05-2016
		EA 201690243 A1	30-06-2016
		EP 3027209 A1	08-06-2016
		JP 2016528232 A	15-09-2016
		KR 20160030581 A	18-03-2016
		PH 12016500182 A1	25-04-2016
		SG 11201600731W A	26-02-2016
		TW 201602133 A	16-01-2016
		US 2015037339 A1	05-02-2015
		US 2018155416 A1	07-06-2018
		UY 35682 A	27-02-2015
		WO 2015017576 A1	05-02-2015
		ZA 201601294 B	31-05-2017
WO 2016168613 A1	20-10-2016	AU 2016249015 A1	09-11-2017
		CA 2982810 A1	20-10-2016
		CN 107771081 A	06-03-2018
		EA 201792298 A1	30-04-2018
		EP 3283519 A1	21-02-2018
		JP 2018512435 A	17-05-2018

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2019/020330

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
		KR 20170135967 A	08-12-2017
		US 2016304595 A1	20-10-2016
		WO 2016168613 A1	20-10-2016