Blockade of CD40 ligation or a CD40L gene knockout can retard atherosclerotic plaque development in atherosclerosis-prone mice (Mach F *et al.*, *Nature* 394: 200-203, 1998; and Lutgens E *et al.*, *Nat Med* 11: 1313-1316, 1999). CD40 ligation on immune cells with stimulating anti-CD40 antibodies has been reported to induce the expression of Sema4D (CD100) (Kumanogoh A *et al.*, *Immunity* 13: 621-31, 2000).

[0005] SEMA4D has been implicated in various processes such as enhancing the immune response, angiogenesis, epithelial morphogenesis and bone remodeling (Kruger RP *et al.*, *Nat Rev Mol Cell Biol* 6: 789-800, 2005; Pasterkamp R.J., *Nat Rev Neurosci* 13:605-618, 2012; and Kang S *et al.*, *Seminars in Cell & Dev Biol* 24:163-171, 2013). Lymphocytes, macrophages, endothelial cells and platelets in atherosclerotic plaques express plexin-B1 on their membrane surface, a high affinity receptor for Sema4D (Basile JR *et al.*, *Moll Cell Biol* 25: 6889-6898, 2005; Delaire S *et al.*, *J Immunol* 166: 4348-4354, 2001; and Chabbert-de Ponnat I *et al.*, *Int Immunol* 17: 439-447, 2005). Sema4D expressed by T cells or released from the T cell membrane can, therefore, have certain effects on cells located in atherosclerotic plaques (Basile JR *et al.*, *Mol. Cell Biol* 25: 6889-6898, 2005; Delaire S *et al.*, *J Immunol* 166: 4348-4354, 2001; and Chabbert-de Ponnat I *et al.*, *Int Immunol* 17: 439-447, 2005). Previous studies revealed pro-angiogenic activity of Sema4D on endothelial cells *in vitro* and *in vivo* (Conrotto P *et al.*, *Blood* 105: 4321-4329, 2005; and Basil JR *et al.*, *Cancer Res* 64: 5212-5224, 2004.). Moreover, high level of expression of Sema4D was observed in several squamous cell carcinomas suggesting a critical role of Sema4D in tumor-induced angiogenesis *in vivo* (Basile JR *et al.*, *Proc Natl Acad Sci USA* 103: 9017-9022, 2006.). Thus, Sema4D can affect plaque growth by, *e.g.*, promoting the neovascularization process occurring in atheroma. In fact, one study revealed that deleting the Sema4D gene in atherosclerosis-prone ApoE deficient (ApoE-/-) mice delayed the growth of atherosclerotic plaques and neovascularization in the plaque area, suggesting that blocking the Sema4D signal during the progression phase of atherosclerosis can prevent the plaque growth and neovascularization (Yukawa K *et al.*, *Int. J Mol. Med.* 26: 39-44, 2010).

BRIEF SUMMARY OF THE INVENTION

[0006] Methods for using semaphorin 4D binding molecules for treatment of atherosclerosis are disclosed herein. According to aspects of the invention illustrated herein, there is provided a use of an effective amount of an isolated binding molecule which specifically

binds to Semaphorin-4D (SEMA4D) in the preparation of a medicament for inhibiting, suppressing, reducing or delaying growth of atherosclerotic plaques in a subject having atherosclerosis. Also disclosed is a method for reducing, inhibiting, suppressing and/or delaying atherosclerotic

plaque formation in a subject having atherosclerosis including administering to the subject an effective amount of an isolated binding molecule which specifically binds to and blocks the activity of semaphorin 4D (SEMA4D). In certain ~~embodiments~~examples, methods for inhibiting, suppressing, reducing or delaying growth of atherosclerotic plaques in a subject having atherosclerosis, comprising administering to the subject an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D) are provided. In certain embodiments, the binding molecule inhibits SEMA4D interaction with its receptor. In certain embodiments, the receptor is Plexin-B1 or Plexin-B2. In certain embodiments, the binding molecule inhibits SEMA4D-mediated Plexin-B1 signal transduction. In certain embodiments of any of the aforementioned methods, the subject has cardiovascular disease. In certain embodiments, the cardiovascular disease is selected from the group consisting of coronary heart disease (also ischemic heart disease or coronary artery disease), cardiomyopathy, hypertensive heart disease, heart failure, cor pulmonale, cardiac dysrhythmias, inflammatory heart disease endocarditis, inflammatory cardiomegaly, myocarditis, valvular heart disease, cerebrovascular disease, peripheral arterial disease, congenital heart disease, rheumatic heart disease, and a combination thereof. In certain embodiments of any of the aforementioned methods, the isolated binding molecule specifically binds to the same SEMA4D epitope as a reference monoclonal antibody VX15/2503 or 67. In certain embodiments of any of the aforementioned methods, the isolated binding molecule competitively inhibits a reference monoclonal antibody VX15/2503 or 67 from specifically binding to SEMA4D. In certain embodiments of any of the aforementioned methods, the isolated binding molecule comprises an antibody or antigen-binding fragment thereof. In certain embodiments of any of the aforementioned methods, the antibody or antigen-binding fragment thereof comprises a variable heavy chain (VH) comprising VHCDRs 1-3 comprising SEQ ID NOs 6, 7, and 8, respectively, and a variable light chain (VL) comprising VLCDRs 1-3 comprising SEQ ID NOs 14, 15, and 16, respectively. In certain embodiments of any of the aforementioned methods, the VH and VL comprise, respectively, SEQ ID NO: 9 and SEQ ID NO: 17 or SEQ ID NO: 10 and SEQ ID NO: 18. In certain ~~embodiments~~ examples of any of the aforementioned methods, the method further includes inhibiting, delaying, reducing or delaying neovascularization around the atherosclerotic plaques.

[0007] Methods for inhibiting, delaying, reducing or delaying neovascularization in a subject having atherosclerosis, comprising administering to the subject an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D) are also ~~provided~~disclosed. In another aspect, there is provided use of an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D), in the preparation of a medicament for inhibiting, reducing or delaying neovascularization in a subject having atherosclerosis. In certain embodiments, the binding molecule inhibits SEMA4D interaction with its receptor. In certain embodiments, the receptor is Plexin-B1. In certain embodiments, the binding molecule inhibits SEMA4D-mediated Plexin-B1 signal transduction. In certain embodiments of any of the aforementioned methods, the subject has cardiovascular disease. In certain embodiments, the cardiovascular disease is selected from the group consisting of coronary heart disease (also ischemic heart disease or coronary artery disease), cardiomyopathy, hypertensive heart disease, heart failure, cor pulmonale, cardiac dysrhythmias, inflammatory heart disease endocarditis, inflammatory cardiomegaly, myocarditis, valvular heart disease, cerebrovascular disease, peripheral arterial disease, congenital heart disease, rheumatic heart disease, and a combination thereof. In certain embodiments of any of the aforementioned methods, the isolated binding molecule specifically binds to the same SEMA4D epitope as a reference monoclonal antibody VX15/2503 or 67. In certain embodiments of any of the aforementioned methods, the isolated binding molecule competitively inhibits a reference monoclonal antibody VX15/2503 or 67 from specifically binding to SEMA4D. In certain embodiments of any of the aforementioned methods, the isolated binding molecule comprises an antibody or antigen-binding fragment thereof. In certain embodiments of any of the aforementioned methods, the antibody or antigen-binding fragment thereof comprises a variable heavy chain (VH) comprising VHCDRs 1-3 comprising SEQ ID NOs 6, 7, and 8, respectively, and a variable light chain (VL) comprising VLCDRs 1-3 comprising SEQ ID NOs 14, 15, and 16, respectively. In certain embodiments of any of the aforementioned methods, the VH and VL comprise, respectively, SEQ ID NO: 9 and SEQ ID NO: 17 or SEQ ID NO: 10 and SEQ ID NO: 18.

[0008] Methods for treating a subject having atherosclerosis, comprising: administering to a subject determined to have atherosclerotic plaques an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D), thereby inhibiting,

delaying, reducing or delaying growth of atherosclerotic plaques are also ~~provided~~disclosed. In another aspect, there is provided use of an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D) in the preparation of a medicament for inhibiting, delaying, reducing or delaying growth of atherosclerotic plaques in a subject determined to have atherosclerotic plaques. In certain embodiments, the determination is provided by analysis of an image or a sample from the patient. In certain embodiments of any of the aforementioned methods, the patient is

[0009] According to aspects illustrated herein, ~~there is provided~~also disclosed is a method of reducing neovascularization in a subject having atherosclerosis including administering to the subject an effective amount of an isolated binding molecule which specifically binds to semaphorin 4D (SEMA4D).

BRIEF DESCRIPTION OF THE DRAWINGS/FIGURES

[0010] FIGURES 1A: Inhibited development of atherosclerotic plaques in anti-Sema4D antibody-treated spontaneously hyperlipidemic (SHL) mice with ApoE-deficiency. FIG. 1A shows a quantitative measurement of the sudanophilic area in lipid deposits of atherosclerotic plaque development in SHL mice treated with anti-SEMA4D antibody or isotype control for 12 weeks starting at 14 weeks of age.

[0011] FIGURES 1B and 1C: Reduced neovascularization in plaques of anti-Sema4D antibody-treated SHL mice. FIG. 1B shows neovascularization visualized by a quantitative measurement of isolectin B4 staining of endothelial cells in plaques of anti-Sema4D antibody-treated SHL mice and control antibody-treated SHL mice. FIG. 1C shows endothelial cells in plaques of anti-Sema4D antibody-treated SHL mice and control antibody-treated SHL mice visualized with immunohistochemistry using CD31 antibodies.

DETAILED DESCRIPTION OF THE INVENTION

I. Definitions

[0012] It is to be noted that the term "a" or "an" entity refers to one or more of that entity; for example, "an anti-SEMA4D antibody" is understood to represent one or more anti-SEMA4D antibodies. As such, the terms "a" (or "an"), "one or more," and "at least one" can be used interchangeably herein.

[0013] As used herein, the term “atherosclerosis” refers to the disorder of the larger arteries that underlies most coronary artery disease, aortic aneurysm, and arterial disease of the lower extremities, and also plays a major role in cerebrovascular disease. Atherosclerosis is a leading cause of death in the United States, both above and below age 65 in both sexes. E.L. Bierman, "Atherosclerosis and Other Forms of Arteriosclerosis," Ch. 208, p. 1 106 in Harrison's Principles of Internal Medicine. 13th edition, eds. KJ. Isselbacher, *et al.* (McGraw-Hill, Inc. NY 1994). Atherosclerosis can affect any artery in the body, including

WHAT IS CLAIMED IS:

1. ~~A method~~Use of an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D) in the preparation of a medicament for inhibiting, suppressing, reducing or delaying growth of atherosclerotic plaques in a subject having atherosclerosis, ~~comprising administering to the subject an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D)~~.

2. The ~~method~~ use of claim 1, wherein the binding molecule inhibits SEMA4D interaction with its receptor.

3. The ~~method~~ use of claim 2, wherein the receptor is Plexin-B1 or Plexin-B2.

4. The ~~method~~ use of claim 1, wherein the binding molecule inhibits SEMA4D-mediated Plexin-B1 signal transduction.

5. The ~~method~~ use of claim 1, wherein the subject has cardiovascular disease.

6. The ~~method~~ use of claim 5, wherein the cardiovascular disease is selected from the group consisting of coronary heart disease (also ischemic heart disease or coronary artery disease), cardiomyopathy, hypertensive heart disease, heart failure, cor pulmonale, cardiac dysrhythmias, inflammatory heart disease endocarditis, inflammatory cardiomegaly, myocarditis, valvular heart disease, cerebrovascular disease, peripheral arterial disease, congenital heart disease, rheumatic heart disease, and a combination thereof.

7. The ~~method~~ use of claim 1, wherein the isolated binding molecule specifically binds to the same SEMA4D epitope as a reference monoclonal antibody VX15/2503 or 67.

8. The ~~method~~ use of claim 1, wherein the isolated binding molecule competitively inhibits a reference monoclonal antibody VX15/2503 or 67 from specifically binding to SEMA4D.

9. The ~~method~~ use of claim 1, wherein the isolated binding molecule comprises an antibody or antigen-binding fragment thereof.

10. The ~~method~~ use of claim 9, wherein the antibody or antigen-binding fragment thereof comprises a variable heavy chain (VH) comprising VHCDRs 1-3 comprising SEQ ID NOs 6, 7, and 8, respectively, and a variable light chain (VL) comprising VLCDRs 1-3 comprising SEQ ID NOs 14, 15, and 16, respectively.

11. The ~~method~~ use of claim 10, wherein the VH and VL comprise, respectively, SEQ ID NO: 9 and SEQ ID NO: 17 or SEQ ID NO: 10 and SEQ ID NO: 18.

12. The ~~method~~ use of claim 1, wherein the method further includes inhibiting, delaying, reducing or delaying neovascularization around the atherosclerotic plaques.

13. ~~A method for inhibiting, delaying, reducing or delaying neovascularization in a subject having atherosclerosis, comprising administering to the subject~~Use of an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D), in the preparation of a medicament for inhibiting, reducing or delaying neovascularization in a subject having atherosclerosis.

14. The ~~method~~ use of claim 13, wherein the binding molecule inhibits SEMA4D interaction with its receptor.

15. The ~~method~~ use of claim 14, wherein the receptor is Plexin-B1.

16. The ~~method~~ use of claim 13, wherein the binding molecule inhibits SEMA4D-mediated Plexin-B1 signal transduction.

17. The ~~method~~ use of claim 13, wherein the subject has cardiovascular disease.

18. The ~~method~~ use of claim 17, wherein the cardiovascular disease is selected from the group consisting of coronary heart disease (also ischemic heart disease or coronary artery disease), cardiomyopathy, hypertensive heart disease, heart failure, cor pulmonale, cardiac dysrhythmias, inflammatory heart disease endocarditis, inflammatory cardiomegaly, myocarditis, valvular heart disease, cerebrovascular disease, peripheral arterial disease, congenital heart disease, rheumatic heart disease, and a combination thereof.

19. The ~~method~~ use of claim 13, wherein the isolated binding molecule specifically binds to the same SEMA4D epitope as a reference monoclonal antibody VX15/2503 or 67.

20. The ~~method~~ use of claim 13, wherein the isolated binding molecule competitively inhibits a reference monoclonal antibody VX15/2503 or 67 from specifically binding to SEMA4D.

21. The ~~method~~ use of claim 13, wherein the isolated binding molecule comprises an antibody or antigen-binding fragment thereof.

22. The ~~method~~ use of claim 21, wherein the antibody or antigen-binding fragment thereof comprises a variable heavy chain (VH) comprising VHCDRs 1-3 comprising SEQ ID NOs 6, 7, and 8, respectively, and a variable light chain (VL) comprising VLCDRs 1-3 comprising SEQ ID NOs 14, 15, and 16, respectively.

23. The ~~method~~ use of claim 22, wherein the VH and VL comprise, respectively, SEQ ID NO: 9 and SEQ ID NO: 17 or SEQ ID NO: 10 and SEQ ID NO: 18.

24. ~~A method for treating a subject having atherosclerosis, comprising: administering to a subject determined to have atherosclerotic plaques~~ Use of an effective amount of an isolated binding molecule which specifically binds to Semaphorin-4D (SEMA4D)~~, thereby~~ in the preparation of a medicament for inhibiting, delaying, reducing or delaying growth of atherosclerotic plaques in a subject determined to have atherosclerotic plaques.