

Additional therapeutic agents that target VEGF or immune checkpoints are needed.

SUMMARY

5 In an aspect, there is provided a multifunctional protein molecule comprising at least one decorin molecule having at least 95% sequence identity to SEQ ID NO: 7 linked to an antibody molecule selected from the group consisting of bevacizumab, ranibizumab, ipilimumab, atezolizumab, avelumab, durvalumab, nivolumab and pembrolizumab.

In another aspect, there is provided a nucleic acid or set of nucleic acids encoding a multifunctional protein molecule as defined above.

10 In another aspect, there is provided a vector or vectors comprising the nucleic acid or set of nucleic acids as defined above.

In another aspect, there is provided a host cell comprising the vector or vectors as defined above.

15 In another aspect, there is provided a multifunctional protein molecule as defined above for use in treating a disorder characterized by angiogenesis or tumor growth.

In another aspect, there is provided the use of the multifunctional protein molecule as defined above in the manufacture of a medicament for the treatment of a disorder characterized by angiogenesis or tumor growth.

20 The present disclosure relates to multifunction protein molecules comprising decorin and uses thereof. In particular, the present disclosure relates to multifunctional protein molecules comprising decorin and a targeting polypeptide such as an antibody and methods of their production and uses thereof.

25 Accordingly, in some examples, the present disclosure provides multifunctional protein molecules comprising at least one decorin molecule or functional portion thereof linked to an antigen binding protein.

In some preferred examples, the antigen binding protein is selected from the group consisting of a VEGF-A antigen binding protein and a checkpoint inhibitor antigen binding protein. In some preferred examples, the checkpoint inhibitor antigen binding protein binds a checkpoint inhibitor protein selected from the group consisting of PD-1, PD-L1, CTLA-4, PD-L2, CD27, CD28, CD40, CD47, CD115, CD122, CD137, OX40, GITR, ICOS, A2AR, B7-H3, 30 B7-H4, BTLA, IDO, KIR, LAG3, NOX2, TIM-3, VISTA, SIGLEC-7,

TIGIT and 4-1BB. In some particularly preferred examples, the checkpoint inhibitor antigen binding protein binds a checkpoint inhibitor protein selected from the group consisting of PD-1, PD-L1, CTLA-4, PD-L2.

5 In some preferred examples, the antigen binding protein is an antibody. In some preferred examples, the antibody is a monoclonal antibody. In some preferred examples, the monoclonal antibody is selected from the group consisting of bevacizumab, ranibizumab, ipilimumab, atezolizumab, avelumab, durvalumab, nivolumab and pembrolizumab.

10 In some preferred examples, the decorin polypeptide is a decorin core protein. In some preferred examples, the decorin core protein comprises a mutation at position 4 of the mature decorin core protein. In some preferred examples, the mutation is a serine to alanine mutation. In some preferred examples, the decorin core protein lacks substantial modification by glycosaminoglycans molecules at position 4 of the mature decorin core protein. In some preferred examples, the fusion protein comprises two or more copies of the decorin polypeptide.

In some preferred examples, the at least a functional portion of a decorin molecule comprises a decorin domain or domains that binds a signaling molecule selected from the group consisting of Transforming Growth Factor- β (TGF- β), Connective Tissue Growth Factor (CTGF), Platelet-Derived Growth Factor (PDGF), Vascular Endothelial Growth Factor Receptor 2 (VEGFR2), Hepatocyte Growth Factor Receptor (HGFR), Insulin-like Growth Factor 1 Receptor (IGF-1R), Epidermal Growth Factor Receptors (EGFRs), myostatin and C1q. In some preferred examples, the TGF- β binding domain comprises amino acids Asp45-Lys359 of full length endogenous human decorin or amino acids Leu155-Val260 of full length endogenous human decorin. In some preferred examples, the multifunctional protein molecule comprises two or more copies of the at least a functional portion of a decorin molecule.

In some preferred examples, the decorin molecule is operably linked to a heavy chain of the antibody. In some preferred examples, the antigen binding protein is bi-specific. In some preferred examples, the antigen binding protein is multi-specific.

In some preferred examples, the multifunctional protein molecule is a fusion protein. In some preferred examples, the decorin molecule is chemically linked to the antigen binding protein.

In some preferred examples, the present disclosure provides a nucleic acid or set of nucleic acids encoding a multifunctional protein molecule as described above. In some preferred examples, the present disclosure provides a vector or vectors comprising the nucleic acid or set of nucleic acids. In some preferred examples, the present disclosure provides a host cell comprising the vector or vectors.

In some preferred examples, the present disclosure provides methods of inhibiting a target protein and a signaling molecule in a cell, comprising: contacting the cell with a multifunctional protein molecule, nucleic acid molecule, or vector as described above under conditions such that at least one activity of the target protein and at least one activity of a signaling protein are inhibited in the cell, wherein the target protein is selected from the group consisting of VEGF-1, PD-1, PD-L1, CTLA-4, PD-L2, CD27, CD28, CD40, CD47, CD115, CD122, CD137, OX40, GITR, ICOS, A2AR, B7-H3, B7-H4, BTLA, IDO, KIR, LAG3,

NOX2, TIM-3, VISTA, SIGLEC-7, TIGIT and 4-1BB. In some preferred examples, the cell is in vitro or in vivo. In some preferred examples, the cell is in a subject. In some preferred examples, the contacting results in inhibition of an activity selected from the group

consisting of angiogenesis, PD-1 activity, PD-L1 activity, CTLA-4 activity, PD-L2 activity, CD27 activity, CD28 activity, CD40 activity, CD47 activity, CD115 activity, CD122 activity, CD137 activity, OX40 activity, GITR activity, ICOS activity, A2AR activity, B7-H3 activity, B7-H4 activity, BTLA activity, IDO activity, KIR activity, LAG3 activity, NOX2 activity, TIM-3 activity, VISTA activity, SIGLEC-7 activity, TIGIT activity and 4-1BB activity. In some preferred examples, the cancer is selected from the group consisting of lung cancer, colorectal cancer, liver cancer, breast cancer, kidney cancer, cervical cancer, ovarian cancer, and glioblastoma. In some preferred examples, the signaling protein is selected from the group consisting of Transforming Growth Factor- β (TGF- β), Connective Tissue Growth Factor (CTGF), Platelet-Derived Growth Factor (PDGF), Vascular Endothelial Growth Factor Receptor 2 (VEGFR2), Hepatocyte Growth Factor Receptor (HGFR), Insulin-like Growth Factor 1 Receptor (IGF-1R), various epidermal growth factor receptors (EGFRs), myostatin and C1q. In some preferred examples, the signaling protein is Transforming Growth Factor- β (TGF- β).

In some preferred examples, the present disclosure provides methods of treating a disorder characterized by angiogenesis or tumor growth, comprising: administering a multifunctional protein molecule, nucleic acid molecule, or vector as described above to a subject under conditions such that angiogenesis or tumor growth is inhibited in the subject. In some preferred examples, the tumor is selected from the group consisting of lung cancer, colorectal cancer, liver cancer, breast cancer, kidney cancer, cervical cancer, ovarian cancer, and glioblastoma.

DESCRIPTION OF THE FIGURES

FIG. 1 is a schematic diagram of a fusion protein of the present invention.

FIG. 2 is a map of an expression construct of the present invention.

FIG. 3 is a map of an expression construct of the present invention.

FIG. 4 is a SDS-PAGE gel of media from the pooled CHO cell line expressing the bevacizumab fusion protein. Lane 10. Molecular weight standards. Lane 11. Non-reduced media sample. Lane 12. Reduced media sample.

CLAIMS

What is claimed is:

5

1. A multifunctional protein molecule comprising at least one decorin molecule having at least 95% sequence identity to SEQ ID NO: 7 linked to an antibody molecule selected from the group consisting of bevacizumab, ranibizumab, ipilimumab, atezolizumab, avelumab, durvalumab, nivolumab and pembrolizumab.

10

2. The multifunctional protein molecule of claim 1, wherein the decorin molecule comprises a mutation at position 4 as compared to wild-type decorin.

15

3. The multifunctional protein molecule of claim 2, wherein the mutation is a serine to alanine mutation.

4. The multifunctional protein molecule of claim 2, wherein the decorin molecule lacks modification by glycosaminoglycans molecules at position 4 of the decorin molecule.

20

5. The multifunctional protein molecule of claim 1, wherein the multifunctional protein molecule comprises two or more copies of the decorin molecule.

25

6. The multifunctional protein molecule of claim 1, wherein the decorin molecule comprises a decorin domain or domains that binds a signaling molecule selected from the group consisting of Transforming Growth Factor- β (TGF- β), Connective Tissue Growth Factor (CTGF), Platelet-Derived Growth Factor (PDGF), Vascular Endothelial Growth Factor Receptor 2 (VEGFR2), Hepatocyte Growth Factor Receptor (HGFR), Insulin-like Growth Factor 1 Receptor (IGF-1R), Epidermal Growth Factor Receptors (EGFRs), myostatin and C1q.

7. The multifunctional protein molecule of claim 6, wherein the TGF- β binding domain comprises amino acids Asp45-Lys359 of full length endogenous human decorin or amino acids Leu155-Val260 of full length endogenous human decorin.
- 5 8. The multifunctional protein molecule of claim 6, wherein the multifunctional protein molecule comprises two or more copies of the decorin molecule.
9. The multifunctional protein molecule of claim 1, wherein the decorin molecule is operably linked to a heavy chain of the antibody.
- 10 10. The multifunctional protein molecule of claim 1, wherein the multifunctional protein molecule is a fusion protein.
11. The multifunctional protein molecule of claim 1, wherein the decorin molecule is
15 chemically linked to the antigen binding protein.
12. A nucleic acid or set of nucleic acids encoding a multifunctional protein molecule of claim 1.
- 20 13. A vector or vectors comprising the nucleic acid or set of nucleic acids of claim 12.
14. A host cell comprising the vector or vectors of claim 13.
15. Multifunctional protein molecule of claim 1 for use in treating a disorder characterized by
25 angiogenesis or tumor growth.
16. Use of the multifunctional protein molecule of claim 1 in the manufacture of a medicament for the treatment of a disorder characterized by angiogenesis or tumor growth.

17. The use of claim 16, wherein the tumor is selected from the group consisting of lung cancer, colorectal cancer, liver cancer, breast cancer, kidney cancer, cervical cancer, ovarian cancer, and glioblastoma.