CLAIMS

1. An isolated fibronectin based scaffold (FBS) protein that specifically binds to a target, wherein the FBS is a non-naturally occurring FBS, and wherein the FBS is linked at its C-terminus to a moiety consisting of the amino acid sequence PmXn, wherein P is proline, X is any amino acid, m is an integer that is at least 1 and n is 0 or an integer that is at least 1, and wherein the PmXn moiety provides an enhanced property to the FBS protein relative to the FBS protein that is not linked to the PmXn moiety.

2. The isolated FBS protein of claim 1, wherein the moiety consists of P (m is 1 and n is 0).

3. The isolated FBS protein of claim 1, wherein the moiety consists of PP (m is 2 and n is 0).

4. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-150.

5. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-10.

6. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-5.

7. The isolated FBS protein of claim 1, wherein the moiety consists of PI, PC, PID, PIE, PIDK (SEQ ID NO: 61), PIEK (SEQ ID NO: 63), PIDKP (SEQ ID NO: 65), PIEKP (SEQ ID NO: 67), PIDKPS (SEQ ID NO: 69), PIEKPS (SEQ ID NO: 71), PIDKPC (SEQ ID NO: 73), PIEKPC (SEQ ID NO: 75), PIDKPSQ (SEQ ID NO: 77), PIEKPSQ (SEQ ID NO: 79), PIDKPCQ (SEQ ID NO: 81), PIEKPCQ (SEQ ID NO: 83), PHHHHHH (SEQ ID NO: 87) or PCHHHHHH (SEQ ID NO: 86).

8. The isolated FBS protein of any one of claims 1-7, wherein the FBS protein is an Fn3 protein.

9. The isolated FBS protein of claim 8, wherein the Fn3 protein is a ^{10}Fn3 protein.

10. The isolated FBS protein of claim 9, wherein the ^{10}Fn3 protein is a human ^{10}Fn3 protein.

11. The isolated FBS protein of any one of claims 1-10, comprising an amino acid sequence that is at least 50% identical to SEQ ID NO: 1.

12. The isolated FBS protein of any one of claims 1-11, comprising at least one amino acid mutation in at least one loop or one scaffold region.

13. The isolated FBS protein of any one of claims 1-12, comprising the amino acid sequence VSDVPRDLEVVAA$(X)_u$LLISW$(X)_v$YRITY$(X)_w$FTV$(X)_x$ATISGL$(X)_y$YTITVY A$(X)_z$ISINYRT (SEQ ID NO: 9), wherein $(X)_u$, $(X)_v$, $(X)_w$, $(X)_x$, $(X)_y$ and $(X)_z$ consist of the wild-type amino acid sequence (SEQ ID NO: 1) or comprise at least one amino acid difference with the corresponding wild-type sequence, and the sequence optionally comprises from 1-10 scaffold, N-terminal and/or C-terminal mutations.

14. The isolated FBS protein of any one of claims 1-13, comprising an amino acid sequence that is at least 50% identical to SEQ ID NO: 1, and wherein the C-terminal amino acid residue of the FBS protein is fused to a moiety consisting of PmXn, wherein the FBS protein binds specifically to a target with a Kd of less than 500 nM, which target is not bound by a wild-type ^{10}Fn3 molecule comprising SEQ ID NO: 1.

15. The isolated FBS protein of claim 14, comprising an amino acid sequence that is at least 70% identical to SEQ ID NO: 1.

16. The isolated FBS protein of any one of claims 1-15, wherein the moiety consisting of PmXn is linked to the C-terminal amino acid residue of the FBS protein through a peptide bond.

17. The isolated FBS protein of any one of claims 1-16, wherein PmXn is P or PC.

18. The isolated FBS protein of any one of claims 1-17, comprising an amino acid sequence that is at least 60% identical to SEQ ID NO: 1, and wherein the C-terminal amino acid residue of the FBS protein is linked to a proline through a peptide bond, and wherein the FBS protein binds specifically to a target with a Kd of less than 500 nM, which target is not bound by a wild-type ^{10}Fn3 molecule comprising SEQ ID NO: 1.

19. The isolated FBS protein of any of claims 1-18, wherein at least one X of PmXn is a cysteine.

20. The isolated FBS protein of claim 19, wherein the cysteine is conjugated to a heterologous moiety.

21. The isolated FBS protein of claim 20, wherein the heterologous molecule is a detectable moiety.

22. The isolated FBS protein of claim 20 or 21, wherein the heterologous molecule is a drug moiety and the drug moiety and the FBS form an FBS-drug conjugate.

23. The isolated FBS protein of any one of claims 1-22, wherein the enhanced property conferred by the PmXn moiety is enhanced stability.

24. The isolated FBS protein of claim 23, wherein enhanced stability is an increase in Tm of at least 1 °C, 2 °C, 3 °C, 4 °C, 5 °C or more.

Claims

1. An isolated fibronectin based scaffold (FBS) protein that specifically binds to Proprotein Convertase Subtilisin Kexin type 9 (PCSK9), wherein the FBS is a non-naturally occurring FBS, and wherein the FBS is linked at its C-terminus to a moiety consisting of the amino acid sequence PmXn, wherein P is proline, X is any amino acid, m is an integer that is at least 1 and n is 0 or an integer that is at least 1, and wherein the PmXn moiety provides an enhanced property to the FBS protein relative to the FBS protein that is not linked to the PmXn moiety.

2. The isolated FBS protein of claim 1, wherein the moiety consists of P (m is 1 and n is 0).

3. The isolated FBS protein of claim 1, wherein the moiety consists of PP (m is 2 and n is 0).

4. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-150.

5. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-10.

6. The isolated FBS protein of claim 1, wherein the moiety consists of PmXn, wherein n is 1-5.

7. The isolated FBS protein of claim 1, wherein the moiety consists of PI, PC, PID, PIE, PIDK (SEQ ID NO: 61), PIEK (SEQ ID NO: 63), PIDKP (SEQ ID NO: 65), PIEKP (SEQ ID NO: 67), PIDKPS (SEQ ID NO: 69), PIEKPS (SEQ ID NO: 71), PIDKPC (SEQ ID NO: 73), PIEKPC (SEQ ID NO: 75), PIDKPSQ (SEQ ID NO: 77), PIEKPSQ (SEQ ID NO: 79), PIDKPCQ (SEQ ID NO: 81), PIEKPCQ (SEQ ID NO: 83), PHHHHHH (SEQ ID NO: 87) or PCHHHHHH (SEQ ID NO: 86).

8. The isolated FBS protein of any one of claims 1-7, wherein the FBS protein is an Fn3 protein.

9. The isolated FBS protein of claim 8, wherein the Fn3 protein is a ^{10}Fn3 protein.

10. The isolated FBS protein of claim 9, wherein the ^{10}Fn3 protein is a human ^{10}Fn3 protein.

11. The isolated FBS protein of any one of claims 1-10, comprising an amino acid sequence that is at least 50% identical to SEQ ID NO: 1.

12. The isolated FBS protein of any one of claims 1-11, comprising at least one amino acid mutation in at least one loop or one scaffold region.

13. The isolated FBS protein of any one of claims 1-12, comprising the amino acid sequence VSDVPRDLEVVAA$(X)_u$LLISW$(X)_v$YRITY$(X)_w$FTV$(X)_x$ATISGL$(X)_y$YTITVY A$(X)_Z$ISINYRT (SEQ ID NO: 9), wherein $(X)_u$, $(X)_v$, $(X)_w$, $(X)_x$, $(X)_y$ and $(X)_z$ consist of the wild-type amino acid sequence (SEQ ID NO: 1) or comprise at least one amino acid difference with the corresponding wild-type sequence, and the sequence optionally comprises from 1-10 scaffold, N-terminal and/or C-terminal mutations.

14. The isolated FBS protein of any one of claims 1-13, comprising an amino acid sequence that is at least 50% identical to SEQ ID NO: 1,

wherein the C- terminal amino acid residue of the FBS protein is fused to a moiety consisting of PmXn, and

wherein the FBS protein binds specifically to Proprotein Convertase Subtilisin Kexin type 9 (PCSK9) with a Kd of less than 500 nM, and PCSK9 is not bound by a wild-type ^{10}Fn3 molecule comprising SEQ ID NO: 1.

15. The isolated FBS protein of claim 14, comprising an amino acid sequence that is at least 70% identical to SEQ ID NO: 1.

16. The isolated FBS protein of any one of claims 1-15, wherein the moiety consisting of PmXn is linked to the C-terminal amino acid residue of the FBS protein through a peptide bond.

17. The isolated FBS protein of any one of claims 1-16, wherein PmXn is P or PC.

18. The isolated FBS protein of any one of claims 1-17, comprising an amino acid sequence that is at least 60% identical to SEQ ID NO: 1,

wherein the C- terminal amino acid residue of the FBS protein is linked to a proline through a peptide bond, and

wherein the FBS protein binds specifically to Proprotein Convertase Subtilisin Kexin type 9 (PCSK9) with a Kd of less than 500 nM, and PCSK9 is not bound by a wild-type ^{10}Fn3 molecule comprising SEQ ID NO: 1.

19. The isolated FBS protein of any of claims 1-18, wherein at least one X of PmXn is a cysteine.

20. The isolated FBS protein of claim 19, wherein the cysteine is conjugated to a heterologous moiety.

21. The isolated FBS protein of claim 20, wherein the heterologous molecule is a detectable moiety.

22. The isolated FBS protein of claim 20 or 21, wherein the heterologous molecule is a drug moiety and the drug moiety and the FBS form an FBS-drug conjugate.

23. The isolated FBS protein of any one of claims 1-22, wherein the enhanced property conferred by the PmXn moiety is enhanced stability.

24. The isolated FBS protein of claim 23, wherein enhanced stability is an increase in Tm of at least 1°C, 2°C, 3°C, 4°C, 5°C or more.