one aspect, there is provided a polypeptide comprising an anti-CX3CR1 (anti-CX3 chemokine receptor 1) immunoglobulin single variable domain, wherein said anti-CX3CR1 immunoglobulin single variable domain consists essentially of four framework regions (FR1, FR2, FR3 and FR4) and three complementary determining regions (CDR1, CDR2 and CDR3) and wherein said CDR3 has the amino acid sequence of Asp-Pro-Arg-Arg-Gly-Trp-Asp-Thr-Arg-Tyr (SEQ ID NO: 186) and wherein said polypeptide comprises a CDR1, CDR2 and CDR3 having the amino acid sequences set forth in:

- SEQ ID No: 213, 221 and 186, respectively; or
- SEQ ID No: 141, 162 and 186, respectively; or
- ~~SEQ ID No: 141, 163 and 187, respectively; or~~
- SEQ ID No: 141, 164 and 186, respectively; or
- SEQ ID No: 141, 166 and 186, respectively; or
- SEQ ID No: 141, 167 and 186, respectively; or
- ~~SEQ ID No: 141, 167 and 189, respectively; or~~
- ~~SEQ ID No: 141, 168 and 186, respectively; or~~
- ~~SEQ ID No: 141, 168 and 187, respectively; or~~
- ~~SEQ ID No: 141, 169 and 190, respectively; or~~
- ~~SEQ ID No: 141, 170 and 186, respectively; or~~
- ~~SEQ ID No: 141, 171 and 186, respectively; or~~
- ~~SEQ ID No: 141, 174 and 186, respectively; or~~
- ~~SEQ ID No: 141, 175 and 187, respectively; or~~
- ~~SEQ ID No: 142, 165 and 188, respectively; or~~
- ~~SEQ ID No: 142, 173 and 188, respectively; or~~
- ~~SEQ ID No: 143, 164 and 186, respectively; or~~
- ~~SEQ ID No: 144, 172 and 187, respectively; or~~
- ~~SEQ ID No: 145, 172 and 187, respectively; or~~
- ~~SEQ ID No: 141, 214 and 186, respectively; or~~
- ~~SEQ ID No: 141, 215 and 186, respectively; or~~
- ~~SEQ ID No: 141, 216 and 186, respectively; or~~
- ~~SEQ ID No: 141, 217 and 186, respectively; or~~
- ~~SEQ ID No: 141, 218 and 186, respectively; or~~
- ~~SEQ ID No: 141, 219 and 186, respectively; or~~
- ~~SEQ ID No: 141, 220 and 186, respectively; or~~
- SEQ ID No: 213, 214 and 186, respectively.

In a further aspect, there is provided use of a polypeptide as defined above for the manufacture of a medicament for the treatment, prevention or alleviation of a disease, disorder or condition, in a human being, wherein the disease or disorder is selected from cardio- and cerebrovascular atherosclerotic disorders, peripheral artery disease, restenosis, diabetic nephropathy, glomerulonephritis, human crescentic glomerulonephritis, IgA nephropathy, membranous nephropathy, lupus nephritis, vasculitis including Henoch-Schonlein purpura and Wegener's granulomatosis, rheumatoid arthritis, osteoarthritis, allograft rejection, systemic sclerosis, neurodegenerative disorders and demyelinating disease, multiple sclerosis (MS), Alzheimer's disease, pulmonary diseases such as COPD, asthma,neuropathic pain, inflammatory pain, or cancer.

~~In a further aspect, there is provided use of polypeptide as defined above in the manufacture of a medicament for the treatment or prevention of CX3CR1-associated disease or disorder.~~

In a further aspect, there is provided a diagnostic kit comprising a polypeptide as defined above, or the use thereof

In a further aspect, there is provided a polypeptide comprising the amino acid sequence of SEQ ID NO: 225.

In a further aspect, there is provided a polypeptide comprising the amino acid sequence of SEQ ID NO: 226.

In a further aspect, there is provided a polypeptide comprising the amino acid sequence of SEQ ID NO: 227.

In a further aspect, there is provided a polypeptide comprising the amino acid sequence of SEQ ID NO: 258.

In a further aspect, there is provided a polypeptide comprising the amino acid sequence of SEQ ID NO: 261.

In a further aspect, there is provided a pharmaceutical composition comprising (i) a polypeptide as defined above, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

The polypeptides of the present disclosure are used for the prevention, treatment, alleviation and/or diagnosis of CX3CR1-associated diseases, disorders or conditions, in particular cardiovascular diseases, such as atherosclerosis.

The present disclosure may provide:

Embodiment 1: An immunoglobulin comprising an antigen-binding domain comprising three complementarity determining regions CDR1, CDR2 and CDR3, wherein said immunoglobulin binds to human CX3CR1 and is capable of blocking the binding of human fractalkine to human CX3CR1.

Embodiment 2: A polypeptide comprising one or more anti-CX3CR1 immunoglobulin single variable domain, wherein said polypeptide is capable of blocking the binding of human fractalkine to human CX3CR1.

Embodiment 3: A polypeptide according to embodiment 2, wherein said anti-CX3CR1 immunoglobulin single variable domain consists essentially of four framework regions (FR1, FR2, FR3 and FR4) and three complementary determining regions (CDR1, CDR2 and CDR3).

Embodiment 4: A polypeptide according to embodiment 3, wherein said anti-CX3CR1 immunoglobulin single variable domain has the structure FR1 - CDR1 - FR2 - CDR2 - FR3 - CDR3 - FR4.

Embodiment 5: A polypeptide according to any one of embodiments 2 to 4, wherein said anti-CX3CR1 immunoglobulin single variable domain is an antibody domain.

Embodiment 6: A polypeptide according to embodiment 5, wherein said anti-CX3CR1 immunoglobulin single variable domain is a VH, VL, VHH, camelized

Claims

1. A polypeptide comprising an anti-CX3CR1 (anti-CX3 chemokine receptor 1) immunoglobulin single variable domain, wherein said anti-CX3CR1 immunoglobulin single variable domain consists essentially of four framework regions (FR1, FR2, FR3 and FR4) and three complementary determining regions (CDR1, CDR2 and CDR3) and wherein said CDR3 has the amino acid sequence of Asp-Pro-Arg-Arg-Gly-Trp-Asp-Thr-Arg-Tyr (SEQ ID NO: 186) and wherein said polypeptide comprises a CDR1, CDR2 and CDR3 having the amino acid sequences set forth in:

- SEQ ID No: 213, 221 and 186, respectively; or
- SEQ ID No: 141, 162 and 186, respectively; or
- ~~SEQ ID No: 141, 163 and 187, respectively; or~~
- SEQ ID No: 141, 164 and 186, respectively; or
- SEQ ID No: 141, 166 and 186, respectively; or
- SEQ ID No: 141, 167 and 186, respectively; or
- ~~SEQ ID No: 141, 167 and 189, respectively; or~~
- ~~SEQ ID No: 141, 168 and 186, respectively; or~~
- ~~SEQ ID No: 141, 168 and 187, respectively; or~~
- ~~SEQ ID No: 141, 169 and 190, respectively; or~~
- ~~SEQ ID No: 141, 170 and 186, respectively; or~~
- ~~SEQ ID No: 141, 171 and 186, respectively; or~~
- ~~SEQ ID No: 141, 174 and 186, respectively; or~~
- ~~SEQ ID No: 141, 175 and 187, respectively; or~~
- ~~SEQ ID No: 142, 165 and 188, respectively; or~~
- ~~SEQ ID No: 142, 173 and 188, respectively; or~~
- ~~SEQ ID No: 143, 164 and 186, respectively; or~~
- ~~SEQ ID No: 144, 172 and 187, respectively; or~~

~~- SEQ ID No: 145, 172 and 187, respectively; or~~

~~- SEQ ID No: 141, 214 and 186, respectively; or~~

~~- SEQ ID No: 141, 215 and 186, respectively; or~~

~~- SEQ ID No: 141, 216 and 186, respectively; or~~

~~- SEQ ID No: 141, 217 and 186, respectively; or~~

~~- SEQ ID No: 141, 218 and 186, respectively; or~~

~~- SEQ ID No: 141, 219 and 186, respectively; or~~

~~- SEQ ID No: 141, 220 and 186, respectively; or~~

- SEQ ID No: 213, 214 and 186, respectively.

2. The polypeptide according to claim 1, wherein the polypeptide comprises a CDR1, CDR2 and CDR3 having amino acid sequences set forth in: SEQ ID NO's: 141, 164 and 186, respectively, SEQ ID NO's: 141, 162 and 186, respectively, SEQ ID NO's: 213, 214 and 186 respectively, or SEQ ID NO's: 213, 221 and 186 respectively.

3. The polypeptide according to any one of claims 1 to 2, wherein said anti-CX3CR1 immunoglobulin single variable domain is a VHH domain comprising the sequence set forth in:

~~a) the amino acid sequence of SEQ ID NO: 3;~~

~~b) an amino acid sequence that has at least 90% amino acid identity with the amino acid sequences of SEQ ID NO: 3; or~~

~~c~~a) an amino acid sequence of any one of SEQ ID NO: 1, 3-5~~-48~~, 7, 9, 10, 13, 19, 21, 23, 24, 121-~~140~~ 130 , 138-140 or ~~—~~ 222-224.

6. The polypeptide according to claim 4 or 5, wherein said half-life extending moiety consists of an anti-albumin immunoglobulin single variable domain.

7. The polypeptide according to claim 6, wherein the immunoglobulin single variable domain is selected from a VHH domain, a humanized VHH domain, a camelized VH domain, a domain antibody, a single domain antibody and/or "dAb"s.

8. The polypeptide according to claim 7, wherein the anti-albumin immunoglobulin single variable domain comprises a sequence selected from any one of SEQ ID NO's: 230-232.

9. The polypeptide according to claim 1, wherein said polypeptide further comprises a second immunoglobulin single variable domain.

10. The polypeptide according to claim 9 wherein said second immunoglobulin single variable domain comprises a second anti-CX3CR1 immunoglobulin single variable domain.

~~11. The polypeptide according to claim 9, wherein said first and second immunoglobulin single variable domains are a VH, VL, VHH, camelized VH, or VHH that is optimized for stability, potency, manufacturability and/or similiarity to human framework regions.~~

~~12~~11. The polypeptide according to claim ~~10~~9, wherein said second immunoglobulin single variable domain comprises CDR1, CDR2 and CDR3 amino acid sequences set forth in

- SEQ ID No: 213, 221 and 186, respectively; or
- SEQ ID No: 141, 162 and 186, respectively; or
- ~~SEQ ID No: 141, 163 and 187, respectively; or~~

- SEQ ID No: 141, 164 and 186, respectively; or
- SEQ ID No: 141, 166 and 186, respectively; or
- SEQ ID No: 141, 167 and 186, respectively; or
~~- SEQ ID No: 141, 167 and 189, respectively; or~~
~~- SEQ ID No: 141, 168 and 186, respectively; or~~
~~- SEQ ID No: 141, 168 and 187, respectively; or~~
~~- SEQ ID No: 141, 169 and 190, respectively; or~~
~~- SEQ ID No: 141, 170 and 186, respectively; or~~
~~- SEQ ID No: 141, 171 and 186, respectively; or~~
~~- SEQ ID No: 141, 174 and 186, respectively; or~~
~~- SEQ ID No: 141, 175 and 187, respectively; or~~
~~- SEQ ID No: 142, 165 and 188, respectively; or~~
~~- SEQ ID No: 142, 173 and 188, respectively; or~~
~~- SEQ ID No: 143, 164 and 186, respectively; or~~
~~- SEQ ID No: 144, 172 and 187, respectively; or~~
~~- SEQ ID No: 145, 172 and 187, respectively; or~~
~~- SEQ ID No: 141, 214 and 186, respectively; or~~
~~- SEQ ID No: 141, 215 and 186, respectively; or~~
~~- SEQ ID No: 141, 216 and 186, respectively; or~~
~~- SEQ ID No: 141, 217 and 186, respectively; or~~
~~- SEQ ID No: 141, 218 and 186, respectively; or~~
~~- SEQ ID No: 141, 219 and 186, respectively; or~~
~~- SEQ ID No: 141, 220 and 186, respectively; or~~
- SEQ ID No: 213, 214 and 186, respectively.

~~13~~12. The polypeptide according to claim ~~12~~11, wherein said first and said second immunoglobulin single variable domains comprise the same CDR1, CDR2 and CDR3.

~~14~~13. The polypeptide according to claim ~~13~~12, wherein said first and second immunoglobulin single variable domains comprise a VHH domain having an amino acid sequence set forth in

~~a) the amino acid sequence of SEQ ID NO: 3;~~

~~b) an amino acid sequence that has at least 90% amino acid identity with the amino acid sequences of SEQ ID NO: 3; or~~

~~c~~a) an amino acid sequence of any one of SEQ ID NO: 1, 3-5, 7, 9, 10, 13, 19, 21, 23, 24-~~48~~, 121-~~140~~ 130 or ~~222~~-224~~;~~

~~d) the amino acid sequence of SEQ ID NO: 49;~~

~~e) an amino acid sequence that has at least 95% amino acid identity with the amino acid sequences of SEQ ID NO: 49;~~

~~f) an amino acid sequence of any one of SEQ ID NO: 49-52;~~

~~g) the amino acid sequence of SEQ ID NO: 67;~~

~~h) an amino acid sequence that has at least 90% amino acid identity with the amino acid sequences of SEQ ID NO: 67; or~~

~~i) an amino acid sequence of any one of SEQ ID NO: 53-120~~.

~~15~~14. A polypeptide comprising a first immunoglobulin single variable domain and a second immunoglobulin single variable domain, each comprising CDR1, CDR2 and CDR3 having amino acid sequences set forth in SEQ ID NO's: 141, 164 and 186, or SEQ ID NO's: 141, 162 and 186, or SEQ ID NO's: 213, 214 and 186, or SEQ ID NO's: 213, 221 and 186.

~~16~~15. A polypeptide comprising the amino acid sequence of any one of SEQ ID NO: 225-227 or 257-262.

~~17~~16. A nucleic acid molecule encoding a polypeptide according to any one of claims 1 to ~~16~~15.

~~18~~17. An expression vector comprising a nucleic acid molecule according to claim ~~17~~16.

~~19~~18. A host cell comprising a nucleic acid molecule encoding a polypeptide according to any one of claims 1 to ~~16~~15, wherein said host cell is capable of expressing said polypeptide.

~~20~~19. A pharmaceutical composition comprising (i) a polypeptide according to any one of claims 1 to ~~16~~15, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

~~21~~20. The pharmaceutical composition according to claim ~~17~~16, wherein said pharmaceutical composition is suitable for intravenous or subcutaneous injection in a human being.

~~22~~21. A method of manufacturing a polypeptide according to any one of claims 1 to ~~16~~15, comprising the steps of

- culturing a host cell under conditions that allow expression of a polypeptide according to any one of claims 1 to ~~16~~15, wherein said host cell is carrying an expression vector comprising a nucleic acid molecule, said nucleic acid molecule comprising a region encoding a polypeptide according to any one of claims 1 to ~~16~~15, and wherein said host cell is a prokaryotic or a eukaryotic cell.

~~23~~22. The method according to claim ~~22~~21, further comprising the steps of

- recovering said polypeptide; and
- purifying said polypeptide.

~~24~~23. A polypeptide according to any of claims 1 to ~~16~~15 for use in the treatment, prevention or alleviation of a disease, disorder or condition, in a human being.

~~25~~24. The polypeptide for use according to claim ~~24~~23, wherein the disease, disorder or condition is a CX3CR1-associated disease, disorder or condition.

~~26~~25. The polypeptide for use according to claim ~~25~~24, wherein the disease, disorder or condition is selected from cardio- and cerebrovascular atherosclerotic

disorders, peripheral artery disease, restenosis, diabetic nephropathy, glomerulomephritis, human crescentic glomerulonephritis, IgA nephropathy, membranous nephropathy, lupus nephritis, vasculitis including Henoch-Schonlein purpura and Wegener's granulomatosis, rheumatoid arthritis, osteoarthritis, allograft rejection, systemic sclerosis, neurodegenerative disorders and demyelinating disease, multiple sclerosis (MS), Alzheimer's disease, pulmonary diseases such as COPD, asthma,neuropathic pain, inflammatory pain, or cancer.

~~27~~26. The polypeptide for use according to claim ~~26~~25, wherein the disease, disorder or condition is atherosclerosis.

~~28~~27. Use of a polypeptide according to any of claims 1 to ~~16~~15 for the manufacture of a medicament for the treatment, prevention or alleviation of a disease, disorder or condition, in a human being.

~~29. Use of a polypeptide according to any one of claims 1 to 16 in the manufacture of a medicament for the treatment or prevention of CX3CR1-associated disease or disorder.~~

~~30. The use according to claim 29~~, wherein the disease or disorder is selected from cardio- and cerebrovascular atherosclerotic disorders, peripheral artery disease, restenosis, diabetic nephropathy, glomerulonephritis, human crescentic glomerulonephritis, IgA nephropathy, membranous nephropathy, lupus nephritis, vasculitis including Henoch-Schonlein purpura and Wegener's granulomatosis, rheumatoid arthritis, osteoarthritis, allograft rejection, systemic sclerosis, neurodegenerative disorders and demyelinating disease, multiple sclerosis (MS), Alzheimer's disease, pulmonary diseases such as COPD, asthma,neuropathic pain, inflammatory pain, or cancer.

~~31~~28. The use according to claim ~~29~~27, wherein the disease or disorder is atherosclerosis.

~~32~~29. A diagnostic kit comprising a polypeptide according to any one of claims 1 to ~~16~~15, or the use thereof.

~~33~~30. A diagnostic kit according to claim ~~32~~29, for the diagnosis of at least one of cardio- and cerebrovascular atherosclerotic disorders, peripheral artery disease, restenosis, diabetic nephropathy, glomerulomephritis, human crescentic glomerulonephritis, IgA nephropathy, membranous nephropathy, lupus nephritis, vasculitis including Henoch-Schonlein purpura and Wegener's granulomatosis, rheumatoid arthritis, osteoarthritis, allograft rejection, systemic sclerosis, neurodegenerative disorders and demyelinating disease, multiple sclerosis (MS), Alzheimer's disease, pulmonary diseases such as COPD, asthma,neuropathic pain, inflammatory pain, or cancer.

~~34~~31. A polypeptide comprising the amino acid sequence of SEQ ID NO: 225.

~~35~~32. A polypeptide comprising the amino acid sequence of SEQ ID NO: 226.

~~36~~33. A polypeptide comprising the amino acid sequence of SEQ ID NO: 227.

~~37~~34. A polypeptide comprising the amino acid sequence of SEQ ID NO: 258.

~~38~~35. A polypeptide comprising the amino acid sequence of SEQ ID NO: 261.

~~39~~36. A pharmaceutical composition comprising (i) a polypeptide according to claim ~~35~~31, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

~~40~~37. A pharmaceutical composition comprising (i) a polypeptide according to claim ~~36~~32, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

~~41~~38. A pharmaceutical composition comprising (i) a polypeptide according to claim ~~37~~33, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

~~42~~39. A pharmaceutical composition comprising (i) a polypeptide according to claim ~~38~~34, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.

~~43~~40. A pharmaceutical composition comprising (i) a polypeptide according to claim ~~39~~35, and (ii) a pharmaceutically acceptable carrier, and optionally (iii) a diluent, excipient, adjuvant and/or stabilizer.