

[0008] Also disclosed is a fusion molecule comprising a canine IgG Fc region variant disclosed herein and a polypeptide. In some examples, the canine IgG Fc region variant is covalently attached to the polypeptide (e.g., through a hinge region or a linker). In some instances, the polypeptide is a ligand binding domain of a canine receptor protein, an extracellular domain of a canine receptor protein, or an antigen-binding domain. In some instances, the polypeptide is selected from the ligand binding domain or extracellular domain of canine IL-13R α 1, or IL-13R α 2, canine EPO, canine CTLA4, canine LFA3, canine VEGFR1/VEGFR3, canine IL-1R, canine GLP-1 receptor agonist, and canine Thrombopoietin binding peptide. In some instances, the polypeptide is a scFv, a nanobody, or single domain antibody. In some instances, the IgG Fc region variant is a variant of a canine IgG.B antibody Fc region. In some instances, the IgG Fc region variant is a variant of a canine IgG.A antibody Fc region. In some instances, the IgG Fc region variant is a variant of a canine IgG.C antibody Fc region. In some instances, the IgG Fc region variant is a variant of a canine IgG.D antibody Fc region.

[0008a] In one aspect, the present disclosure refers to a polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises (i) a Tyr at amino acid position 252 of the wild type canine IgG Fc region, and (ii) at least one of the following:

- Asp or Glu at amino acid position 251,
- Asn or Asp at amino acid position 285,
- Asp at amino acid position 286,
- Gln at amino acid position 307,
- Pro at amino acid position 308,
- Asp at amino acid position 315,

Ala or Lys at amino acid position 430,
Lys at amino acid position 433,
Tyr at amino acid position 435, and
His at amino acid position 436,

wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;

(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;

(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and

(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

[0008b] In one aspect, the present disclosure refers to a polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises (i) a Met at amino acid position 252 of the wild type canine IgG Fc region, and (ii) at least one of the following:

Asp or Glu at amino acid position 251,
Asp or Phe at amino acid position 256,
Asn or Asp at amino acid position 285,
Asp at amino acid position 286,
Gln at amino acid position 307,
Pro at amino acid position 308,
Asp at amino acid position 315,
Ala or Lys at amino acid position 430,
Lys at amino acid position 433,
Tyr at amino acid position 435, and
His at amino acid position 436,

wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;

(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;

(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and

(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

[0008c] In one aspect, the present disclosure refers to a polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises an Arg at amino acid position 434 of the wild type canine IgG Fc region, wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;

(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;

(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and

(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

[0009] In one example, the disclosure provides a polypeptide or polypeptides comprising a canine IgG Fc CH2 region variant, the CH2 region variant comprising an amino acid sequence that is at least 75% identical to the sequence set forth in SEQ ID NO:1, and comprises at least one of the following:

an amino acid other than Ile at amino acid position 250,
an amino acid other than Leu at amino acid position 251,
an amino acid other than Arg at amino acid position 252,
an amino acid other than Thr at amino acid position 254,
an amino acid other than Thr at amino acid position 256,
an amino acid other than His at amino acid position 285,
an amino acid other than Thr at amino acid position 286,
an amino acid other than Pro at amino acid position 307,
an amino acid other than Ile at amino acid position 308,
an amino acid other than Glu at amino acid position 309,
an amino acid other than Gln at amino acid position 311, or
an amino acid other than Thr at amino acid position 315.

[0010] In some examples, the polypeptide or polypeptides has/have: (1) increased half-life in a dog than a control polypeptide or control polypeptides, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc CH2 region in place of the IgG Fc CH2

WHAT IS CLAIMED IS:

1. A polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises (i) a Tyr at amino acid position 252 of the wild type canine IgG Fc region, and (ii) at least one of the following:

Asp or Glu at amino acid position 251,
Asn or Asp at amino acid position 285,
Asp at amino acid position 286,
Gln at amino acid position 307,
Pro at amino acid position 308,
Asp at amino acid position 315,
Ala or Lys at amino acid position 430,
Lys at amino acid position 433,
Tyr at amino acid position 435, and
His at amino acid position 436,

wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;
(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;
(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and
(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

2. A polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises (i) a Met at amino acid position 252 of the wild type canine IgG Fc region, and (ii) at least one of the following:

Asp or Glu at amino acid position 251,
Asp or Phe at amino acid position 256,
Asn or Asp at amino acid position 285,
Asp at amino acid position 286,
Gln at amino acid position 307,

Pro at amino acid position 308,
Asp at amino acid position 315,
Ala or Lys at amino acid position 430,
Lys at amino acid position 433,
Tyr at amino acid position 435, and
His at amino acid position 436,

wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;

(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;

(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and

(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

3. A polypeptide comprising a canine IgG Fc region variant, wherein the canine IgG Fc region variant comprises an Arg at amino acid position 434 of the wild type canine IgG Fc region, wherein the amino acid positions are based on EU numbering, wherein the polypeptide has increased binding to canine FcRn than a control polypeptide, wherein the control polypeptide or control polypeptides are identical to the polypeptide or polypeptides except for having the corresponding wild type canine IgG Fc region in place of the IgG Fc region variant, and wherein the wild type canine IgG is selected from the group consisting of:

(a) a canine IgG.A comprising an Fc domain having the amino acid sequence of SEQ ID NO: 9;

(b) a canine IgG.B comprising an Fc domain having the amino acid sequence of SEQ ID NO: 10;

(c) a canine IgG.C comprising an Fc domain having the amino acid sequence of SEQ ID NO: 11; and

(d) a canine IgG.D comprising an Fc domain having the amino acid sequence of SEQ ID NO: 12.

4. The polypeptide of any one of claims 1 to 3, wherein the polypeptide further comprises a binding domain.

5. The polypeptide of claim 4, wherein the binding domain comprises (i) six complementarity determining regions (CDRs) of an immunoglobulin molecule; (ii) a ligand binding domain of a canine receptor protein; (iii) a nanobody; or (iv) an extracellular domain of a canine receptor protein.

6. The polypeptide of claim 4 or claim 5, wherein the binding domain specifically binds to an antigen selected from the group consisting of NGF, TrKA, ADAMTS, IL-1, IL-2, IL-4, IL-4R, Angiotensin type 1 (AT1) receptor, Angiotensin type 2 (AT2) receptor, IL-5, IL-12, IL-13, IL-31, IL-33, CD3, CD20, CD47, CD52, and complement system complex.

7. The polypeptide of any one of claims 1 to 3, further comprising a protein selected from the group consisting of EPO, CTLA4, LFA3, VEGFR1/VEGFR3, IL-1R, IL-4R, GLP-1 receptor agonist, and Thrombopoietin binding peptide.

8. The polypeptide of any one of claims 1 to 7, wherein the polypeptide binds to a canine FcRn at a higher level at an acidic pH than at a neutral pH.

9. A pharmaceutical composition comprising (i) the polypeptide of any one of claims 1 to 8, and (ii) a pharmaceutically acceptable excipient.

10. A nucleic acid or nucleic acids encoding the polypeptide of any one of claims 1 to 8.

11. An expression vector or expression vectors comprising the nucleic acid or nucleic acids of claim 10.

12. A host cell comprising the nucleic acid or nucleic acids of claim 10 or the expression vector or expression vectors of claim 11.

13. A method of making a polypeptide, the method comprising:
- (a) providing a nucleic acid or nucleic acids of claim 10;
 - (b) expressing the nucleic acid or nucleic acids in a host cell culture, thereby producing the polypeptide; and
 - (c) collecting the polypeptide produced in (b) from the host cell culture.
14. The method of claim 13, further comprising formulating the polypeptide as a pharmaceutical formulation.
15. Use of a composition comprising the pharmaceutical composition of claim 9 in the manufacture of a medicament for treating a canine disease or disorder in a dog in need thereof.
16. Use of a composition comprising the pharmaceutical composition of claim 9 in the manufacture of a medicament for preventing a canine disease or disorder in a dog in need thereof.
17. The use of claim 15 or claim 16, wherein the canine disease or disorder is an allergic disease, chronic pain, acute pain, an inflammatory disease, an autoimmune disease, an endocrine disease, a gastrointestinal disease, a cardiovascular disease, a renal disease, a fertility related disorder, an infectious disease, or a cancer.
18. The use of claim 15 or claim 16, wherein the canine disease or disorder is atopic dermatitis, allergic dermatitis, osteoarthritic pain, arthritis, anemia, or obesity.