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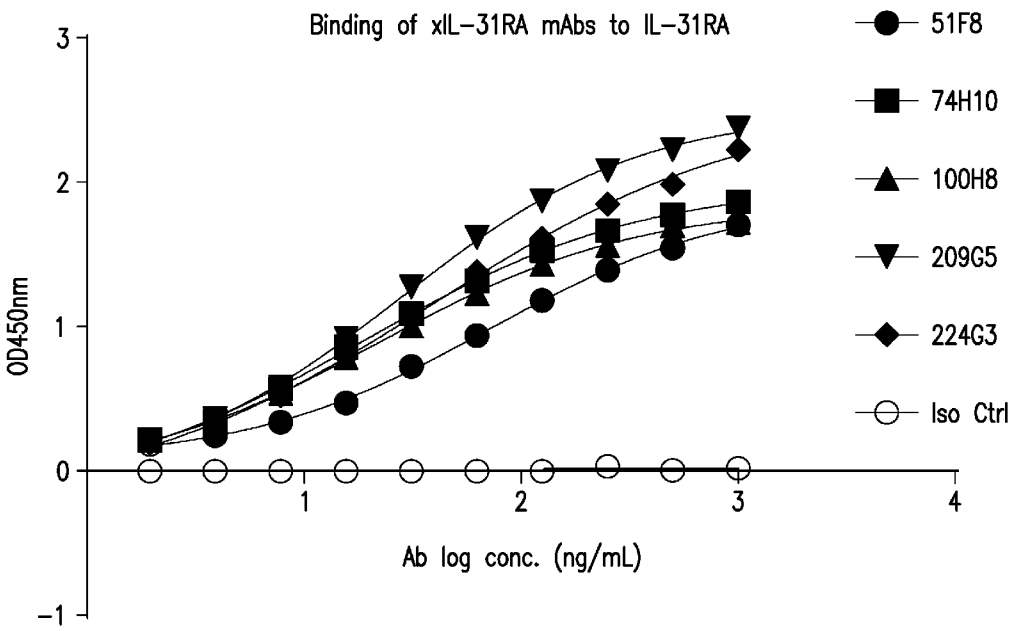


FIG.2A

(57) Abstract: The present invention provides caninized mouse antibodies to canine IL-31 receptor alpha that have a high binding affinity for canine IL-31 receptor alpha, and that can block the binding of canine IL-31 to canine IL-31 receptor alpha. The present invention further provides the use of the antibodies for the treatment of atopic dermatitis in dogs.

[Continued on next page]

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CANINIZED ANTIBODIES TO CANINE INTERLEUKIN-31 RECEPTOR *ALPHA* II

REFERENCE TO SEQUENCE LISTING SUBMITTED ELECTRONICALLY

The instant application contains a Sequence Listing which has been submitted electronically in XML format and is hereby incorporated by reference in its entirety. The XML file, created on December 9, 2022, is named 25365.xml. This sequence listing submitted via EFS-Web is part of the specification and is herein incorporated by reference in its entirety.

CROSS-REFERENCE TO RELATED APPLICATIONS

This application claims priority under 35 U.S.C. § 119(e) of provisional applications U.S. Serial No. 63/341,443, filed on May 13, 2022, U.S. Serial No. 63/290,259 filed on December16, 2021, and U.S. Serial No. 63/290,256, filed on December16, 2021. The subject matter of which are hereby incorporated by reference in their entireties.

FIELD OF THE INVENTION

The present invention relates to antibodies to canine IL-31 receptor *alpha* that have a high binding affinity for canine IL-31 receptor *alpha*, and that can block the binding of canine IL-31 to the canine IL-31 receptor *alpha*. The present invention also relates to use of the antibodies of the present invention in the treatment of atopic dermatitis in dogs.

BACKGROUND OF THE INVENTION

The immune system comprises a network of resident and recirculating specialized cells that function collaboratively to protect the host against infectious diseases and cancer. The ability of the immune system to perform this function depends to a large extent on the biological activities of a group of proteins secreted by leukocytes and collectively referred to as interleukins. Among the well-studied interleukins are four important molecules identified as interleukin-31 (IL-31), interleukin-4 (IL-4), interleukin-13 (IL-13), and interleukin-22 (IL-22). Although IL-4, IL-13, IL-22, and IL-31, are critical cytokines for the development of immune responses that are required for protection against extracellular pathogens (*e.g.*, tissue or lumen dwelling parasites), these cytokines also have been implicated in the pathogenesis of allergic diseases in humans and animals, including atopic dermatitis.

Atopic dermatitis (AD) is a relapsing pruritic and chronic inflammatory skin disease, that is characterized by immune system dysregulation and epidermal barrier abnormalities in humans.

The pathological and immunological attributes of atopic dermatitis have been the subject of extensive investigations [reviewed in Rahman *et al. Inflammation & Allergy-drug target* 10:486-496 (2011) and Harskamp *et al., Seminar in Cutaneous Medicine and Surgery* 32:132-139 (2013)]. Atopic dermatitis is also a common condition in companion animals, especially dogs, where its prevalence has been estimated to be approximately 10-15% of the canine population. The pathogenesis of atopic dermatitis in dogs and cats [*reviewed in* Nuttall *et al., Veterinary Records* 172(8):201-207 (2013)] shows significant similarities to that of atopic dermatitis in man including skin infiltration by a variety of immune cells and $CD4^+$ Th2 polarized cytokine milieu including the preponderance of IL-31, IL-4, and IL-13. In addition, IL-22 has been implicated in the exaggerated epithelial proliferation leading to epidermal hyperplasia that is characteristic of atopic dermatitis.

For example, antibodies against canine IL-31 have been shown to have an effect on pruritus associated with atopic dermatitis in dogs [US 8,790,651 B2; US 10,093,731 B2]. In addition, an antibody against human IL-31 receptor *alpha* (IL-31RA) has been tested and found to have an effect on pruritus associated with atopic dermatitis in humans [Ruzicka, *et al.*, *New England Journal of Medicine*, 376(9),826–835 (2017)].

Pharmaceuticals that have either proven to aid in the treatment of atopic dermatitis and/or have shown promise to do so include: Janus kinase (JAK) inhibitors [*see e.g.,* U.S. 8,133,899; U.S. 8,987,283; WO 2018/108969], spleen tyrosine kinase (SYK) inhibitors [*see e.g.*, U.S. 8,759,366], and antagonists to a chemoattractant receptor-homologous molecule expressed on TH2 cells [*see e.g.,* U.S. 7,696,222, U.S. 8,546,422, U.S. 8,637,541, and U.S. 8,546,422].

However, despite some recent success in treating atopic dermatitis, there remains a need to design alternative and/or better therapies that can address one or more of the symptoms of canine atopic dermatitis.

The citation of any reference herein should not be construed as an admission that such reference is available as "prior art" to the instant application.

SUMMARY OF THE INVENTION

The present invention provides new mammalian antibodies, including caninized murine antibodies, to IL-31 receptor *alpha* (IL-31RA) from canines. In certain embodiments, the mammalian antibodies to canine IL-31 receptor *alpha* (cIL-31RA) are isolated antibodies. In preferred embodiments, the mammalian antibodies or antigen binding fragments thereof bind canine IL-31RA. In more particular embodiments, the mammalian antibodies or antigen binding

fragments also block the binding of canine IL-31RA to canine interleukin-31. In certain embodiments, the mammalian antibodies are antibodies to canine IL-31RA. In more particular embodiments, the mammalian antibodies are caninized antibodies. In even more particular embodiments, the caninized antibodies are caninized murine antibodies to canine IL-31RA.

Accordingly, the present invention provides mammalian antibodies or antigen binding fragments thereof that bind canine interleukin-31 receptor *alpha* and that comprise a heavy chain that comprises a set of three heavy chain complementary determining regions (HCDRs), a CDR heavy 1 (HCDR1), a CDR heavy 2 (HCDR2), and a CDR heavy 3 (HCDR3) in which the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3; the HCDR2 comprises the amino acid sequence of SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, or SEQ ID NO: 13; and the HCDR3 comprises the amino acid sequence of SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, or SEQ ID NO: 23.

In certain embodiments, the mammalian antibody or antigen binding fragment further comprises a light chain that comprises a set of three light chain complementary determining regions (LCDRs) a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3) in which the LCDR1 comprises the amino acid sequence of SEQ ID NO: 30 or SEQ ID NO: 31; the LCDR2 comprises the amino acid sequence of SEQ ID NO: 38, SEQ ID NO: 39, or SEQ ID NO: 40; and the LCDR3 comprises the amino acid sequence of SEQ ID NO: 45.

In particular embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to one or more epitopes comprised by the amino acid sequences of SEQ ID NO: 97, SEQ ID NO: 103, SEQ ID NO: 99, or SEQ ID NO: 102. In more particular embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to two epitopes, one epitope of which is comprised by the amino acid sequence of SEQ ID NO: 97 and the other epitope is comprised by the amino acid sequence of SEQ ID NO: 103. In other embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to two epitopes, one epitope of which is comprised by the amino acid sequence of SEQ ID NO: 99 and the other epitope is comprised by the amino acid sequence of SEQ ID NO: 102.

In particular embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to an epitope comprised by the amino acid sequence of SEQ ID NO: 97. In other embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 103. In still other embodiments, when bound to canine IL-31RA the

mammalian antibody or antigen binding fragment thereof binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 99. In yet other embodiments, when bound to canine IL-31RA the mammalian antibody or antigen binding fragment thereof binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 102.

In related embodiments, when bound to canine IL-31RA, the antibody binds at least one amino acid residue, preferably one to three amino acid residues, more preferably two to five amino acid residues, and/or more preferably three to eight amino acid residues or more within the amino acid sequence of SEQ ID NO: 97 or SEQ ID NO: 103, or both SEQ ID NO: 97 and SEQ ID NO: 103, and/or SEQ ID NO: 99 or SEQ ID NO: 102, or both SEQ ID NO: 99 and SEQ ID NO: 102.

In particular embodiments, the mammalian antibody or antigen binding fragment thereof comprises an HCDR1 that comprises the amino acid sequence of SEQ ID NO: 3, an HCDR2 that comprises the amino acid sequence of SEQ ID NO: 10, and an HCDR3 that comprises the amino acid sequence of SEQ ID NO: 20. In more particular embodiments of this type, the mammalian antibody or antigen binding fragment thereof further comprises a LCDR1 that comprises the amino acid sequence of SEQ ID NO: 30, a LCDR2 that comprises the amino acid sequence of SEQ ID NO: 40, and the LCDR3 that comprises the amino acid sequence of SEQ ID NO: 45.

In other embodiments, the mammalian antibody or antigen binding fragment thereof comprises an HCDR1 that comprises the amino acid sequence of SEQ ID NO: 3, an HCDR2 that comprises the amino acid sequence of SEQ ID NO: 11, and an HCDR3 that comprises the amino acid sequence of SEQ ID NO: 21. In more particular embodiments of this type, the mammalian antibody or antigen binding fragment thereof further comprises a LCDR1 that comprises the amino acid sequence of SEQ ID NO: 30, a LCDR2 that comprises the amino acid sequence of SEQ ID NO: 40, and the LCDR3 that comprises the amino acid sequence of SEQ ID NO: 45.

In still other embodiments, the mammalian antibody or antigen binding fragment thereof comprises an HCDR1 that comprises the amino acid sequence of SEQ ID NO: 3, an HCDR2 that comprises the amino acid sequence of SEQ ID NO: 12, and an HCDR3 that comprises the amino acid sequence of SEQ ID NO: 22. In more particular embodiments of this type, the mammalian antibody or antigen binding fragment thereof further comprises a LCDR1 that comprises the amino acid sequence of SEQ ID NO: 30, a LCDR2 that comprises the amino acid sequence of SEQ ID NO: 38, and the LCDR3 that comprises the amino acid sequence of SEQ ID NO: 45.

In yet other embodiments, the mammalian antibody or antigen binding fragment thereof comprises an HCDR1 that comprises the amino acid sequence of SEQ ID NO: 3, an HCDR2 that

comprises the amino acid sequence of SEQ ID NO: 13, and an HCDR3 that comprises the amino acid sequence of SEQ ID NO: 23. In more particular embodiments of this type, the mammalian antibody or antigen binding fragment thereof further comprises a LCDR1 that comprises the amino acid sequence of SEQ ID NO: 31, a LCDR2 that comprises the amino acid sequence of SEQ ID NO: 39, and the LCDR3 that comprises the amino acid sequence of SEQ ID NO: 45.

In preferred embodiments, the antibody and antigen binding fragment thereof bind canine IL-31RA and block the binding of canine IL-31RA to canine interleukin-31. In specific embodiments, the mammalian antibody to canine IL-31RA is a murine antibody. In particular embodiments, the mammalian antibody to canine IL-31RA is a caninized antibody. In more particular embodiments, the caninized antibody to canine IL-31RA is a caninized murine antibody.

The caninized antibodies of the present invention comprise a canine fragment crystallizable region (cFc region). The caninized antibodies of the present invention also comprise a canine light chain constant region. In particular embodiments the canine light chain constant region is a *kappa* canine light chain constant region. In more specific embodiments, the *kappa* canine light chain constant region comprises the amino acid sequence of SEQ ID NO: 127.

Furthermore the caninized antibody or antigen binding fragment thereof can comprise a heavy chain that comprises a cFc region and a hinge region. The hinge region is preferably a canine hinge region. The canine hinge region can comprise a natural occurring: IgG-A hinge region, IgG-B hinge region, IgG-C hinge region, or IgG-D hinge region. Alternatively, the hinge region is a corresponding modified canine hinge region. In particular embodiments, the hinge region is the IgG-A hinge region comprising an amino acid sequence comprising at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 112. In other embodiments, the hinge region is the IgG-B hinge region comprising an amino acid sequence comprising at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 113. In still other embodiments, the hinge region is the IgG-C hinge region comprising an amino acid sequence comprising at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 114. In yet other embodiments, the hinge region is a modified IgG-D hinge region comprising the amino acid sequence of SEQ ID NO: 115.

Similarly, the canine Fc region can be an IgG-A, IgG-B, IgG-C, an IgG-D or modifications thereof. In particular embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-Bm. In certain embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-A that comprises an amino acid sequence that has at

least 90%, 95%, 98%, 99% or 100% identity with the amino acid sequence of SEQ ID NO: 116. In other embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-B that comprises an amino acid sequence that has at least 90%, 95%, 98%, 99% or 100% identity with the amino acid sequence of SEQ ID NO: 110. In still other embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-C that comprises an amino acid sequence that has at least 90%, 95%, 98%, 99% or 100% identity with the amino acid sequence of SEQ ID NO: 117. In yet other embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-D that comprises an amino acid sequence that has at least 90%, 95%, 98%, 99% or 100% identity with the amino acid sequence of SEQ ID NO: 118. In still other embodiments, a caninized antibody or antigen binding fragment thereof comprises an IgG-Bm that comprises an amino acid sequence that has at least 90%, 95%, 98%, 99% or 100% identity with the amino acid sequence of SEQ ID NO: 111, wherein both the aspartic acid residue (D) at position 31 of SEQ ID NO: 110 and the asparagine residue (N) at position 63 of SEQ ID NO: 110, remain substituted by an alanine residue (A) in the sequence of IgG-Bm.

In particular embodiments, the caninized antibody or antigen binding fragment thereof comprises the canine IgG-D, but the naturally occurring IgG-D hinge region is replaced by a hinge region comprising an amino acid sequence comprising at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 112. In other embodiments, the caninized antibody comprises a heavy chain that comprises an IgG-D, but the naturally occurring IgG-D hinge region is replaced by a hinge region comprising an amino acid sequence has at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 113. In still other embodiments, the caninized antibody comprises a heavy chain that comprises an IgG-D, but the naturally occurring IgG-D hinge region is replaced by a hinge region comprising an amino acid sequence has at least 90%, 95%, or 100% identity with the amino acid sequence of SEQ ID NO: 114. In yet other embodiments, the caninized antibody comprises a heavy chain that comprises an IgG-D, but the naturally occurring IgG-D hinge region is replaced by a hinge region comprising the amino acid sequence of SEQ ID NO: 115.

In certain embodiments of the compositions, the caninized antibody against canine IL-31RA comprises a heavy chain comprising the amino acid sequence of SEQ ID NO: 79, SEQ ID NO: 80, or SEQ ID NO: 81 and a light chain comprising the amino acid sequence of SEQ ID NO: 82, SEQ ID NO: 83, SEQ ID NO: 84, or SEQ ID NO: 85. The present invention further provides antigen binding fragments of these caninized antibodies.

In particular embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 84 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80. In other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 84 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81. In yet other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 84 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 79.

In still other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 85 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80. In yet other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 85 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81. In still other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 85 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 79.

In yet other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 83 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80. In still other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 83 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81. In yet other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 83 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 79.

In still other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 82 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80. In yet other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 82 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81. In still other embodiments, the caninized antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO: 82 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 79.

In specific embodiments, when bound to canine IL-31RA, the mammalian antibody (*e.g.*, a caninized antibody) binds to an epitope comprised by the amino acid of SEQ ID NO: 97 or SEQ ID NO: 103, or to both SEQ ID NO: 97 and SEQ ID NO: 103. In particular embodiments, the identification of the epitopes is based on chemical crosslinking and mass spectrometry detection. In related embodiments, when bound to canine IL-31RA, the mammalian antibody

binds at least one amino acid residue, preferably one to three amino acid residues, more preferably two to five amino acid residues, and/or more preferably three to eight amino acid residues or more within the amino acid sequence of SEQ ID NO: 97 or SEQ ID NO: 103, or within both SEQ ID NO: 97 and SEQ ID NO: 103.

In particular embodiments, the mammalian antibody that binds to SEQ ID NO: 97, binds to the tyrosine residue at position 94 of SEQ ID NO. 2, *i.e.* Y_{94}. In other embodiments, the mammalian antibody binds to the lysine residue at position 102 of SEQ ID NO. 2, *i.e.* K_{102}. In still other embodiments, the mammalian antibody binds to the lysine residue at position 112 of SEQ ID NO. 2, *i.e.* K_{112}. In yet other embodiments, the mammalian antibody binds to binds to the tyrosine residue at position 94 of SEQ ID NO. 2, and the lysine residue at position 102 of SEQ ID NO. 2. In still other embodiments, the mammalian antibody binds to the lysine residue at position 102 of SEQ ID NO. 2, and the lysine residue at position 112 of SEQ ID NO. 2. In yet other embodiments, the mammalian antibody binds to binds to the tyrosine residue at position 94 of SEQ ID NO. 2, and the lysine residue at position 112 of SEQ ID NO. 2. In still other embodiments, the mammalian antibody binds to the tyrosine residue at position 94, binds to the lysine residue at position 102 of SEQ ID NO. 2, and the lysine residue at position 112 of SEQ ID NO. 2.

In related embodiments, the mammalian antibody that binds to SEQ ID NO: 103, binds to the arginine residue at position 183 of SEQ ID NO. 2, *i.e.* R_{183}. In other embodiments, the mammalian antibody binds to the serine residue at position 193 of SEQ ID NO. 2, *i.e.* S_{193}. In still other embodiments, the mammalian antibody binds to the threonine residue at position 202 of SEQ ID NO. 2, *i.e.* T_{202}. In yet other embodiments, the mammalian antibody binds to binds to the arginine residue at position 183 of SEQ ID NO. 2, and the serine residue at position 193 of SEQ ID NO. 2. In still other embodiments, the mammalian antibody binds to the serine residue at position 193 of SEQ ID NO. 2, and the threonine residue at position 202 of SEQ ID NO. 2. In yet other embodiments, the mammalian antibody binds to binds to the arginine residue at position 183 of SEQ ID NO. 2, and the threonine residue at position 202 of SEQ ID NO. 2. In still other embodiments, the mammalian antibody binds to the arginine residue at position 183 of SEQ ID NO. 2, binds to the serine residue at position 193 of SEQ ID NO. 2, and the threonine residue at position 202 of SEQ ID NO. 2. The present invention further provides antigen binding fragments of these mammalian antibodies.

The present invention also provides nucleic acids, including isolated nucleic acids, that encode any of: the sets of 3 HCDRs or 3 LCDRs; the heavy chain variable regions of the

caninized antibodies or antigen binding fragments thereof; the heavy chains of the caninized antibodies or antigen binding fragments thereof, the light chain variable regions of the caninized antibodies or antigen binding fragments thereof, and/or the light chains of the caninized antibodies or antigen binding fragments thereof. The present invention further provides a pair of nucleic acids, wherein one of the pair of nucleic acids comprises a nucleotide sequence that encodes the light chain of a specific caninized antibody of any one of the antibodies of the present invention and the other of the pair of nucleic acids comprises a nucleotide sequence that encodes the heavy chain of that (said) specific caninized antibody. The present invention further provides expression vectors that comprise such pairs of nucleic acids, or alternatively individual nucleic acids of the present invention. In addition, the present invention provides pairs of expression vectors, wherein one of the pair of expression vectors comprises a nucleic acid comprising a nucleotide sequence that encodes the light chain of a specific caninized antibody of any one of the antibodies of the present invention, and the other of the pair of expression vectors comprises a nucleic acid comprising a nucleotide sequence that encodes the heavy chain of that (said) specific caninized antibody.

Accordingly, the present invention further provides nucleic acids that encode a set of the three heavy chain complementary determining regions (CDRs), a CDR heavy 1 (HCDR1), a CDR heavy 2 (HCDR2), and a CDR heavy 3 (HCDR3) of a mammalian antibody (including a caninized antibody) of the present invention. In more specific embodiments, the nucleic acid encodes an HCDR1 that comprises the amino acid sequence of SEQ ID NO: 3; the HCDR2 that comprises the amino acid sequence of SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, or SEQ ID NO: 13; and the HCDR3 that comprises the amino acid sequence of SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, or SEQ ID NO: 23; or any combination thereof.

The present invention further provides nucleic acids that encodes a set of the three light chain complementary determining regions (CDRs), a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3) of a mammalian antibody (including a caninized antibody) or an antigen binding fragment thereof of the present invention. In more specific embodiment of this type, the nucleic acid encodes a LCDR1 that comprises the amino acid sequence of SEQ ID NO: 30 or SEQ ID NO: 31; a LCDR2 that comprises the amino acid sequence of SEQ ID NO: 38, SEQ ID NO: 39, or SEQ ID NO: 40; and a LCDR3 that comprises the amino acid sequence of SEQ ID NO: 45; or any combination thereof.

The present invention further provides nucleic acids that encode any of the heavy chains of a mammalian antibody (including a caninized antibody) or an antigen binding fragment

thereof of the present invention. The present invention also provides nucleic acids that encode any of the light chains of a mammalian antibody (including a caninized antibody) or an antigen binding fragment thereof of the present invention. In addition, the present invention provides expression vectors that comprise and can express one or more of the nucleic acids of the present invention, and host cells that comprise one or more of such expression vectors.

The present invention further provides pharmaceutical compositions that comprise the caninized antibodies and antigen binding fragments thereof of the present invention along with a pharmaceutically acceptable carrier and/or diluent. The present invention further provides pharmaceutical compositions that comprise a nucleic acid of the present invention, along with a pharmaceutically acceptable carrier and/or diluent, and/or an expression vector that comprise one or more of the nucleic acids of the present invention, along with a pharmaceutically acceptable carrier and/or diluent.

The present invention also provides methods of treating atopic dermatitis comprising administering one of the aforesaid pharmaceutical compositions to an animal subject that has atopic dermatitis. In particular embodiments, the animal subject is a canine. The present invention also provides methods of aiding in blocking pruritus associated with atopic dermatitis in an animal subject, comprising administering to an animal subject in need thereof of a therapeutically effective amount of a pharmaceutical composition of the present invention. In particular embodiments, the animal subject is a canine.

In addition, the present invention provides methods of producing a caninized antibody or antigen binding fragment thereof that binds canine IL-31RA. In particular embodiments, the method includes culturing a host cell comprising one or more expression vectors that encode and express the light chain of a caninized antibody of the present invention and the heavy chain of that caninized antibody in a culture medium under conditions in which the nucleic acid is expressed, thereby producing a polypeptide comprising the light chain of a caninized antibody of the present invention, and/or the heavy chain of that caninized antibody. The polypeptides are then recovered from the host cell or culture medium. In certain embodiments, the polypeptides comprising the light chain of a caninized antibody of the present invention and the polypeptides comprising the heavy chain of that caninized antibody are combined with each under conditions that are conducive to form a caninized antibody.

These and other aspects of the present invention will be better appreciated by reference to the following Brief Description of the Drawings and the Detailed Description.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the binding of IL-31 to IL-31RA. The extracellular domain (ECD) of canine IL-31RA was tested for its ability to bind to canine IL-31. The results indicate that IL-31RA ECD binds in a dose-dependent manner to biotinylated canine IL-31 with an EC50 of 0.55 ng/ml.

Figures 2A-2B show the binding of xIL-31RA monoclonal antibodies (mABS) to IL-31RA. The selected mouse mAbs were tested for their reactivity to canine IL-31RA. The results indicate that the selected mouse mAbs bind to canine IL-31RA in a dose-dependent manner. All of the 10 mouse monoclonal antibodies have strong binding reactivity to canine IL-31RA. Figure 2A depicts mouse mAbs: 51F8 (•), 74H10 (◙), 100H8(▲), 209G5 (▼), 224G3(♦), and Iso control (o). Figure 2B depicts mouse mAbs: 55B3 (•), 65G9 (◙), 85C10 (▲), 218D9 (▼), 227E7(♦), and Iso control (o).

Figure 3 shows the blocking of the binding of IL-31 to IL-31RA by monoclonal antibodies (mABS) to IL-31RA. The selected mouse mAbs (anti-canine IL-31RA) were tested for their ability to block the binding of IL-31 with IL-31RA/OSMR by Flow Cytometry. The FACS result indicates that the ten mouse mAbs can block the binding of IL-31 with the IL-31RA/OSMR complex presented on CHO- IL-31RA/OSMR cells. Antibodies 51F8, 74H10, 100H8, 209G5, and 218D9 exhibited superior blocking activity.

Figure 4 shows the induction of STAT-3 phosphorylation by IL-31. Ba/f3-OI cells expressing the IL-31 receptor complex were tested for IL-31-induced STAT-3 phosphorylation. The results indicate that STAT-3 phosphorylation was induced by IL 31 in the Baf3-OI cells (◙) in a dose-dependent manner, implying that: (i) the canine IL-31 receptor complex is successfully expressed on the cell surface; (ii) that the binding of canine IL-31 to the IL-31 receptor can stimulate the endogenous STAT3 phosphorylation; and (iii) then initiate its downstream signaling pathway. Ba/f3 cells (o) were used as the control.

Figures 5A-5B show the inhibition of IL-31-mediated STAT-3 phosphorylation in Ba/f3-OI cells by the selected xIL-31RA antibodies. The results indicate that the selected mAbs inhibit IL-31 mediated STAT-3 phosphorylation in a dose-dependent manner in Ba/f3-OI cells. Figure 5A depicts mouse mAbs 209G5 (◙), 218D9 (▲), 85C10 (▼), and IL-31 protein (♦). Figure 5B depicts mouse mAbs 100H8 (⬣), 74H10 (◙), 85C10 (▲), 51F8 (▼), and cIL-31 protein (♦).

Figures 6A-6E provides the epitopes on canine IL-31RA for the antibodies 100H8, 51F8, 218D9, 85C10, and 224G3, respectively. Figure 6A depicts the epitope for 100H8; the epitope

comprises the amino acid sequences of SEQ ID NO: 97 (within SEQ ID NO: 119) and SEQ ID NO: 103 (within SEQ ID NO: 120), respectively. Figure 6B depicts the epitope for 51F8; the epitope comprises comprises the amino acid sequences of SEQ ID NO: 98 (within SEQ ID NO: 121) and SEQ ID NO: 100 (within SEQ ID NO: 122). Figure 6C depicts the epitope for 218D9; the epitope comprises the amino acid sequences of SEQ ID NO: 104 (within SEQ ID NO: 123) and SEQ ID NO: 105 (within SEQ ID NO: 124), respectively. Figure 6D depicts the epitope for 85C10; the epitope comprises the amino acid sequences of SEQ ID NO: 106 (within SEQ ID NO: 125) and SEQ ID NO: 108 (within SEQ ID NO: 126), respectively. Figure 6E depicts the epitope for 224G3; the epitope comprises the amino acid sequence of SEQ ID NO: 109 (also within SEQ ID NO: 126). The position of binding residues of the amino acid sequence of SEQ ID NO: 2 for the respective epitopes on the cIL-31R ECD antigen are also denoted.

Figures 7A-7E provides plots for the binding activity of the identified murine-canine chimeric or caninized antibodies to canine IL-31RA. The results indicate that the caninized antibodies have similar binding affinity as their corresponding parental antibodies (as represented by the murine-canine chimeric antibodies). Figure 7A depicts the binding plots for monoclonal 51F8 antibodies: Chimeric 51F8 (•) c51F8VH3VL6 (◙), c51F8VH3VL7 (▲), c51F8VH4VL6 (▼), and c51F8VH4VL7 (♦); and the iso control (o). Figure 7B depicts the binding plots for monoclonal 100H8 antibodies: Chimeric 100H8 (•), c100H8VH5VL4 (◙), and c100H8VH7VL4 (▲). Figure 7C depicts the binding plots for monoclonal 85C10 antibodies: Chimeric 85C10 (•), c85C10VH3VL2 (◙), and c85C10VH1VL2(▲). Figure 7D depicts the binding plots for monoclonal 218D9 antibodies: Chimeric 218D9 (•), c218D9VH3VL2 (◙), c218D9VH3VL3 (▲), c218D9VH4VL2 (▼), and c218D9VH4VL3 (♦); and the iso control (o). Figure 7E depicts the binding plots for monoclonal 224G3 antibodies: m224G3 Chim (•), c224G3VH2VL2 (◙), and c224G3VH2VL3(▲). The EC50 is provided in the Tables

The term "Chimeric" before the antibody number signifies that the antibody is a murine-canine chimeric antibody, *e.g.*, Chimeric 218D9 or Chimeric 51F8. In addition, an "m" before the antibody number followed by a "Chim" signifies that the antibody is a murine-canine chimeric antibody, *e.g.*, m224G3 Chim. The lower case "c" before the antibody number signifies that it is a caninized antibody, *e.g.*, c218D9VH4VL2.

Figure 8 shows the blocking of the binding of IL-31 to IL-31RA by the inhibition of the IL-31-mediated STAT-3 phosphorylation in Ba/f3-OI cells. The results indicate that the

caninized 218D9 antibodies can inhibit IL-31 mediated STAT-3 phosphorylation in a dose-dependent manner in Ba/f3-OI cells, and that the constructs c218D9VH3VL3 and c218D9VH4VL3 have the same inhibitory activity as the parental mouse-canine chimeric 218D9 antibody: Chimeric 218D9 (•), c218D9VH3VL2 (◙), c218D9VH3VL3 (▲), c218D9VH4VL2 (▼), and c218D9VH4VL3 (♦); and the IL-31 only control (o).

DETAILED DESCRIPTION OF THE INVENTION

In response to need for better therapies for atopic dermatitis, the present invention provides formulations and methodology that can achieve a significant effect on the skin inflammation associated with atopic dermatitis.

ABBREVIATIONS

Throughout the detailed description and examples of the invention the following abbreviations will be used:

ADCC	Antibody-dependent cellular cytotoxicity
CDC	Complement-dependent cyotoxicity
CDR	Complementarity determining region in the immunoglobulin variable regions, defined using the Kabat numbering system
EC50	concentration resulting in 50% efficacy or binding
ELISA	Enzyme-linked immunosorbant assay
FR	Antibody framework region: the immunoglobulin variable regions excluding the CDR regions
IC50	concentration resulting in 50% inhibition
IgG	Immunoglobulin G
Kabat	An immunoglobulin alignment and numbering system pioneered by Elvin A. Kabat [*Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (1991)]
mAb	Monoclonal antibody (also Mab or MAb)
V region	The segment of IgG chains which is variable in sequence between different antibodies.
VH	Immunoglobulin heavy chain variable region
VL	Immunoglobulin light chain variable region

DEFINITIONS

So that the invention may be more readily understood, certain technical and scientific terms are specifically defined below. Unless specifically defined elsewhere in this document, all other technical and scientific terms used herein have the meaning commonly understood by one of ordinary skill in the art to which this invention belongs.

As used herein, including the appended claims, the singular forms of words such as "a," "an," and "the," include their corresponding plural references unless the context clearly dictates otherwise.

"Administration" and "treatment", as it applies to an animal, *e.g.*, a canine subject, cell, tissue, organ, or biological fluid, refers to contact of an exogenous pharmaceutical, therapeutic, diagnostic agent, or composition to the animal *e.g.*, a canine subject, cell, tissue, organ, or biological fluid. Treatment of a cell encompasses contact of a reagent to the cell, as well as contact of a reagent to a fluid, where the fluid is in contact with the cell.

"Administration" and "treatment" also mean *in vitro* and *ex vivo* treatments, *e.g.*, of a cell, by a reagent, diagnostic, binding compound, or by another cell. The term "subject" includes any organism, preferably an animal, more preferably a mammal (*e.g.*, canine or feline) and most preferably a canine.

"Treat" or "treating" means to administer a therapeutic agent, such as a composition containing any of the antibodies of the present invention, internally or externally to *e.g.*, a canine subject or patient having one or more symptoms, or being suspected of having a condition, for which the agent has therapeutic activity. Typically, the agent is administered in an amount effective to alleviate and/or ameliorate one or more disease/condition symptoms in the treated subject or population, whether by inducing the regression of or inhibiting the progression of such symptom(s) by any clinically measurable degree. The amount of a therapeutic agent that is effective to alleviate any particular disease/condition symptom (also referred to as the "therapeutically effective amount") may vary according to factors such as the disease/condition state, age, and weight of the patient (*e.g.*, canine), and the ability of the pharmaceutical composition to elicit a desired response in the subject. Whether a disease/condition symptom has been alleviated or ameliorated can be assessed by any clinical measurement typically used by veterinarians or other skilled healthcare providers to assess the severity or progression status of that symptom. While an embodiment of the present invention (*e.g.*, a treatment method or article of manufacture) may not be effective in alleviating the target disease/condition symptom(s) in every subject, it should alleviate the target disease/condition symptom(s) in a statistically significant number of subjects as determined by any statistical test known in the art such as the

Student's t-test, the chi^2-test, the U-test according to Mann and Whitney, the Kruskal-Wallis test (H-test), Jonckheere-Terpstra-test and the Wilcoxon-test.

"Treatment," as it applies to a human, veterinary (*e.g.*, canine), or research subject, refers to therapeutic treatment, as well as research and diagnostic applications. "Treatment" as it applies to a human, veterinary (*e.g.*, canine), or research subject, or cell, tissue, or organ, encompasses contact of the antibodies of the present invention to *e.g.*, a canine or other animal subject, a cell, tissue, physiological compartment, or physiological fluid.

As used herein, the term "canine" includes all domestic dogs, Canis lupus familiaris or Canis familiaris, unless otherwise indicated.

As used herein, the term "feline" refers to any member of the *Felidae* family. Members of this family include wild, zoo, and domestic members, including domestic cats, pure-bred and/or mongrel companion cats, show cats, laboratory cats, cloned cats, and wild or feral cats

As used herein the term "canine frame" refers to the amino acid sequence of the heavy chain and light chain of a canine antibody other than the hypervariable region residues defined herein as CDR residues. With regard to a caninized antibody, in the majority of embodiments the amino acid sequences of the native canine CDRs are replaced with the corresponding foreign CDRs (*e.g.*, those from a mouse or rat antibody) in both chains. Optionally the heavy and/or light chains of the canine antibody may contain some foreign non-CDR residues, *e.g.*, so as to preserve the conformation of the foreign CDRs within the caninized antibody, and/or to modify the Fc function, as exemplified below and/or disclosed in U.S. 10,106,607 B2, hereby incorporated by reference herein in its entirety.

The "Fragment crystallizable region" abbreviated as "Fc" or as used interchangeably "Fc region" corresponds to the CH3-CH2 portion of an antibody that interacts with cell surface receptors called Fc receptors. The canine fragment crystallizable region (cFc region) of each of the four canine IgGs were first described by Tang *et al.* [*Vet. Immunol. Immunopathol.* 80: 259-270 (2001); *see also*, Bergeron *et al.*, *Vet. Immunol. Immunopathol.* 157: 31-41 (2014) and U.S. 10,106,607 B2].

As used herein the canine Fc (cFc) "IgG-Bm" is canine IgG-B Fc comprising two (2) amino acid residue substitutions, D31A and N63A, as in the amino acid sequence of SEQ ID NO: 111 of IgG-B (*see* below) and without the c-terminal lysine ('K"). Both the aspartic acid residue (D) at position 31 of SEQ ID NO: 110 and the asparagine residue (N) at position 63 of SEQ ID NO: 110, are substituted by an alanine residue (A) in IgG-Bm. These two amino acid residue substitutions serve to significantly diminish the antibody-dependent cytotoxicity (ADCC)

and complement-dependent cytotoxicity (CDC) of the naturally occurring canine IgG-B [*see*, U.S. 10,106,607 B2, the contents of which are hereby incorporated by reference in their entirety]. Further amino acid substitutions to the IgG-Bm are also envisioned, which parallel those which can be made in IgG-B and may include amino acid substitutions to favor heterodimer formation in bispecific antibodies. The amino acid sequence of IgG-B, SEQ ID NO: 110 is:

```
1                                                    50
LGGPSVFIFP PKPKDTLLIA RTPEVTCVVV DLDPEDPEVQ ISWFVDGKQM
↳ CH2

51                                                   100
QTAKTQPREE QFNGTYRVVS VLPIGHQDWL KGKQFTCKVN NKALPSPIER

101                                                  150
TISKARGQAH QPSVYVLPPS REELSKNTVS LTCLIKDFFP PDIDVEWQSN
      ↳ CH3

151                                                  200
GQQEPESKYR TTPPQLDEDG SYFLYSKLSV DKSRWQRGDT FICAVMHEAL

201          215
HNHYTQKSLS HSPGK
```

The amino acid sequence of IgG-Bm, SEQ ID NO: 111, is provided below.

```
LGGPSVFIFPPKPKDTLLIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGT
YRVVSVLPIGHQDWLKGKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVS
LTCLIKDFFPPDIDVEWQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAV
MHEALHNHYTQESLSHSPG
```

As used herein, a "substitution of an amino acid residue" with another amino acid residue in an amino acid sequence of an antibody for example, is equivalent to "replacing an amino acid residue" with another amino acid residue and denotes that a particular amino acid residue at a specific position in the amino acid sequence has been replaced by (or substituted for) by a different amino acid residue. Such substitutions can be particularly designed *i.e.*, purposefully replacing an alanine with a serine at a specific position in the amino acid sequence by *e.g.*, recombinant DNA technology. Alternatively, a particular amino acid residue or string of amino acid residues of an antibody can be replaced by one or more amino acid residues through more natural selection processes *e.g.*, based on the ability of the antibody produced by a cell to bind to a given region on that antigen, *e.g.*, one containing an epitope or a portion thereof, and/or for the

antibody to comprise a particular CDR that retains the same canonical structure as the CDR it is replacing. Such substitutions/replacements can lead to “variant” CDRs and/or variant antibodies.

As used herein, the term "antibody" refers to any form of antibody that exhibits the desired biological activity. An antibody can be a monomer, dimer, or larger multimer. Thus, it is used in the broadest sense and specifically covers, but is not limited to, monoclonal antibodies (including full length monoclonal antibodies), polyclonal antibodies, multi-specific antibodies (*e.g.*, bispecific antibodies), caninized antibodies, fully canine antibodies, chimeric antibodies and camelized single domain antibodies. "Parental antibodies" are antibodies obtained by exposure of an immune system to an antigen prior to modification of the antibodies for an intended use, such as caninization of an antibody for use as a canine therapeutic antibody.

As used herein, antibodies of the present invention that "block" or is “blocking” or is “blocking the binding” of *e.g.*, a canine receptor to its binding partner (ligand), is an antibody that blocks (partially or fully) the binding of the canine receptor to its canine ligand and *vice versa*, as determined in standard binding assays (*e.g.*, BIACore®, ELISA, or flow cytometry).

Typically, an antibody or antigen binding fragment of the invention retains at least 10% of its canine antigen binding activity (when compared to the parental antibody) when that activity is expressed on a molar basis. Preferably, an antibody or antigen binding fragment of the invention retains at least 20%, 50%, 70%, 80%, 90%, 95% or 100% or more of the canine antigen binding affinity as the parental antibody. It is also intended that an antibody or antigen binding fragment of the invention can include conservative or non-conservative amino acid substitutions (referred to as "conservative variants" or "function conserved variants" of the antibody) that do not substantially alter its biologic activity.

"Isolated antibody" refers to the purification status and in such context means the molecule is substantially free of other biological molecules such as nucleic acids, proteins, lipids, carbohydrates, or other material such as cellular debris and growth media. Generally, the term "isolated" is not intended to refer to a complete absence of such material or to an absence of water, buffers, or salts, unless they are present in amounts that substantially interfere with experimental or therapeutic use of the binding compound as described herein.

As used herein, an antibody is said to bind specifically to a polypeptide comprising a given antigen sequence (in this case a portion of the amino acid sequence of canine IL-31RA) if it binds to polypeptides comprising the portion of the amino acid sequence of canine IL-31RA, but does not bind to other canine proteins lacking that portion of the sequence of canine IL-31RA. For example, an antibody that specifically binds to a polypeptide comprising canine

IL-31RA, may bind to a FLAG®-tagged form of canine IL-31RA, but will not bind to other FLAG®-tagged canine proteins.

As used herein, unless otherwise indicated, "antibody fragment" or "antigen binding fragment" refers to antigen binding fragments of antibodies, *i.e.* antibody fragments that retain the ability to bind specifically to the antigen (*e.g.*, canine IL-31RA) bound by the full-length antibody, *e.g.* fragments that retain one or more CDR regions. Examples of antigen binding fragments include, but are not limited to, Fab, Fab', $F(ab')_2$, and Fv fragments; diabodies; linear antibodies; single-chain antibody molecules, *e.g.*, sc-Fv; nanobodies and multispecific antibodies formed from antibody fragments.

An antibody, or binding compound derived from the antigen-binding site of an antibody, binds to its canine antigen, or a variant or mutein thereof, "with specificity" when it has an affinity for that canine antigen or a variant or mutein thereof which is at least ten-times greater, more preferably at least 20-times greater, and even more preferably at least 100-times greater than its affinity for any other canine antigen tested.

As used herein, a "chimeric antibody" is an antibody having the variable domain from a first antibody and the constant domain from a second antibody, where the first and second antibodies are from different species. [U.S. 4,816,567; and Morrison *et al.*, *Proc. Natl. Acad. Sci. USA* 81: 6851-6855 (1984)]. Typically the variable domains are obtained from an antibody from an experimental animal (the "parental antibody"), such as a rodent, and the constant domain sequences are obtained from the animal subject antibodies, *e.g.*, human or canine so that the resulting chimeric antibody will be less likely to elicit an adverse immune response in a human or canine subject respectively, than the parental (*e.g.,* rodent) antibody.

As used herein, the term "caninized antibody" refers to forms of antibodies that contain sequences from both canine and non-canine (*e.g.*, mouse) antibodies. In general, the caninized antibody will comprise substantially all of at least one or more typically, two variable domains in which all or substantially all of the hypervariable loops correspond to those of a non-canine immunoglobulin (*e.g.*, comprising 6 CDRs as exemplified below), and all or substantially all of the framework (FR) regions (and typically all or substantially all of the remaining frame) are those of a canine immunoglobulin sequence. As exemplified herein, a caninized antibody comprises both the three heavy chain CDRs and the three light chain CDRS from a murine (mouse) anti-canine antigen antibody together with a canine frame or a modified canine frame. A modified canine frame comprises one or more amino acids changes as exemplified herein that further optimize the effectiveness of the caninized antibody, *e.g.*, to increase its binding to its

canine antigen and/or its ability to block the binding of that canine antigen to the canine antigen's natural binding partner.

The variable regions of each light/heavy chain pair form the antibody binding site. Thus, in general, an intact antibody has two binding sites. Except in bifunctional or bispecific antibodies, the two binding sites are, in general, the same. Typically, the variable domains of both the heavy and light chains comprise three hypervariable regions, also called complementarity determining regions (CDRs), located within relatively conserved framework regions (FR). The CDRs are usually aligned by the framework regions, enabling binding to a specific epitope. In general, from N-terminal to C-terminal, both light and heavy chains variable domains comprise FR1, CDR1, FR2, CDR2, FR3, CDR3 and FR4. The assignment of amino acids to each domain is, generally, in accordance with the definitions of *Sequences of Proteins of Immunological Interest*, Kabat, *et al.*; National Institutes of Health, Bethesda, Md. ; 5th ed.; NIH Publ. No. 91-3242 (1991); Kabat, *Adv. Prot. Chem.* 32:1-75 (1978); Kabat, *et al.*, *J. Biol. Chem.* 252:6609-6616 (1977); Chothia, *et al.*, *J. Mol. Biol.* 196:901-917 (1987) or Chothia, *et al.*, *Nature* 342:878-883 (1989)].

As used herein, the term "hypervariable region" refers to the amino acid residues of an antibody that are responsible for antigen-binding. The hypervariable region comprises amino acid residues from a "complementarity determining region" or "CDR" (*i.e.* LCDR1, LCDR2 and LCDR3 in the light chain variable domain and HCDR1, HCDR2 and HCDR3 in the heavy chain variable domain). [*See* Kabat *et al. Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (1991), defining the CDR regions of an antibody by sequence; *see also* Chothia and Lesk, *J. Mol. Biol.* 196: 901-917 (1987) defining the CDR regions of an antibody by structure]. As used herein, the term "framework" or "FR" residues refers to those variable domain residues other than the hypervariable region residues defined herein as CDR residues.

There are four known IgG heavy chain subtypes of dog IgG and they are referred to as IgG-A, IgG-B, IgG-C, and IgG-D. The two known light chain subtypes are referred to as *lambda* and *kappa*. In specific embodiments of the invention, besides binding canine IL-31RA, a canine or caninized antibody against its antigen of the present invention optimally has two attributes:

1) Lack of effector functions such as antibody-dependent cytotoxicity (ADCC) and complement-dependent cytotoxicity (CDC), and

2) be readily purified on a large scale using industry standard technologies such as that based on protein A chromatography.

None of the naturally occurring canine IgG isotypes satisfy both criteria. For example, IgG-B can be purified using protein A, but has high level of ADCC activity. On the other hand, IgG-A binds weakly to protein A, but also displays ADCC activity. Moreover, neither IgG-C nor IgG-D can be purified on protein A columns, although IgG-D displays no ADCC activity. (IgG-C has considerable ADCC activity). One way the present invention addresses these issues in certain embodiments is by providing modified canine IgG-B antibodies of the present invention specific to an antigen of the present invention that lack the effector functions such as ADCC and can be easily purified using industry standard protein A chromatography.

As used herein an "antipruritic agent" is a compound, macromolecule, and/or formulation that tends to inhibit, relieve, and/or prevent itching. Antipruritic agents are colloquially referred to as anti-itch drugs.

As used herein an "antipruritic antibody" is an antibody that can act as an antipruritic agent in an animal, including a mammal such as a human, a canine, and/or a feline, particularly with respect to atopic dermatitis. In particular embodiments, the antipruritic antibody binds to specific proteins in the IL-31 signaling pathway, such as IL-31 or its receptor IL-31RA. The binding of the antipruritic antibody to its corresponding antigen (*e.g.*, IL-31 or IL-31RA) inhibits the binding of *e.g.*, IL-31 with IL-31RA, and interferes with and/or prevents the successful signaling of this pathway, and thereby inhibits, relieves, and/or prevents the itching that is otherwise caused by the IL-31 signaling pathway.

"Homology", as used herein, refers to sequence similarity between two polynucleotide sequences or between two polypeptide sequences when they are optimally aligned. When a position in both of the two compared sequences is occupied by the same base or amino acid residue, *e.g.*, if a position in each of two DNA molecules is occupied by adenine, then the molecules are homologous at that position. The percent of homology is the number of homologous positions shared by the two sequences divided by the total number of positions compared ×100. For example, if 6 of 10 of the positions in two sequences are matched or homologous when the sequences are optimally aligned then the two sequences are 60% homologous. Generally, the comparison is made when two sequences are aligned to give maximum percent homology.

Sequence identity refers to the degree to which the amino acids of two polypeptides are the same at equivalent positions when the two sequences are optimally aligned. As used herein one amino acid sequence is 100% "identical" to a second amino acid sequence when the amino acid residues of both sequences are identical. Accordingly, an amino acid sequence is 50%

"identical" to a second amino acid sequence when 50% of the amino acid residues of the two amino acid sequences are identical. The sequence comparison is performed over a contiguous block of amino acid residues comprised by a given protein, *e.g.*, a protein, or a portion of the polypeptide being compared. In particular embodiments, selected deletions or insertions that could otherwise alter the correspondence between the two amino acid sequences are taken into account.

Sequence similarity includes identical residues and nonidentical, biochemically related amino acids. Biochemically related amino acids that share similar properties and may be interchangeable.

"Conservatively modified variants" or "conservative substitution" refers to substitutions of amino acids in a protein with other amino acids having similar characteristics (*e.g.* charge, side-chain size, hydrophobicity/hydrophilicity, backbone conformation and rigidity, etc.), such that the changes can frequently be made without altering the biological activity of the protein. Those of skill in this art recognize that, in general, single amino acid substitutions in non-essential regions of a polypeptide do not substantially alter biological activity [*see, e.g.*, Watson *et al., Molecular Biology of the Gene*, The Benjamin/Cummings Pub. Co., p. 224 (4th Ed.; 1987)]. In addition, substitutions of structurally or functionally similar amino acids are less likely to disrupt biological activity. Exemplary conservative substitutions are set forth in Table A directly below.

TABLE A

EXEMPLARY CONSERVATIVE AMINO ACID SUBSTITUTIONS

Original residue	Conservative substitution
Ala (A)	Gly; Ser
Arg (R)	Lys; His
Asn (N)	Gln; His
Asp (D)	Glu; Asn
Cys (C)	Ser; Ala
Gln (Q)	Asn
Glu (E)	Asp; Gln
Gly (G)	Ala
His (H)	Asn; Gln

Original residue	Conservative substitution
Ile (I)	Leu; Val
Leu (L)	Ile; Val
Lys (K)	Arg; His
Met (M)	Leu; Ile; Tyr
Phe (F)	Tyr; Met; Leu
Pro (P)	Ala; Gly
Ser (S)	Thr
Thr (T)	Ser
Trp (W)	Tyr; Phe
Tyr (Y)	Trp; Phe
Val (V)	Ile; Leu

Function-conservative variants of the antibodies of the invention are also contemplated by the present invention. "Function-conservative variants," as used herein, refers to antibodies or fragments in which one or more amino acid residues have been changed without altering a desired property, such an antigen affinity and/or specificity. Such variants include, but are not limited to, replacement of an amino acid with one having similar properties, such as the conservative amino acid substitutions of Table A above.

"Isolated nucleic acid molecule" means a DNA or RNA of genomic, mRNA, cDNA, or synthetic origin or some combination thereof which is not associated with all or a portion of a polynucleotide in which the isolated polynucleotide is found in nature, or is linked to a polynucleotide to which it is not linked in nature. For purposes of this disclosure, it should be understood that "a nucleic acid molecule comprising" a particular nucleotide sequence does not encompass intact chromosomes. Isolated nucleic acid molecules "comprising" specified nucleic acid sequences may include, in addition to the specified sequences, coding sequences for up to ten or even up to twenty or more other proteins or portions or fragments thereof, or may include operably linked regulatory sequences that control expression of the coding region of the recited nucleic acid sequences, and/or may include vector sequences.

The present invention provides isolated caninized antibodies of the present invention, methods of use of the antibodies in the treatment of a condition *e.g.*, the treatment of atopic dermatitis in canines. In canine, there are four IgG heavy chains referred to as A, B, C, and D. These heavy chains represent four different subclasses of dog IgG, which are referred to as IgG-

A (or IgGA), IgG-B (or IgGB), IgG-C (or IgGC) and IgG-D (or IgGD). Each of the two heavy chains consists of one variable domain (VH) and three constant domains referred to as CH-1, CH-2, and CH-3. The CH-1 domain is connected to the CH-2 domain *via* an amino acid sequence referred to as the "hinge" or alternatively as the "hinge region".

The nucleic acid and amino acid sequences of these four heavy chains were first identified by Tang *et al.* [*Vet. Immunol. Immunopathol.* 80: 259-270 (2001)]. The amino acid and nucleic sequences for these heavy chains are also available from the GenBank data bases. For example, the amino acid sequence of IgGA heavy chain has accession number AAL35301.1, IgGB has accession number AAL35302.1, IgGC has accession number AAL35303.1, and IgGD has accession number (AAL35304.1). Canine antibodies also contain two types of light chains, *kappa* and *lambda*. The DNA and amino acid sequence of these light chains can be obtained from GenBank Databases. For example, the *kappa* light chain amino acid sequence has accession number ABY 57289.1 and the *lambda* light chain has accession number ABY 55569.1.

The known amino acid sequences of the four unmodified canine Fcs are:

cIgG-A [SEQ ID NO: 116]

LGGPSVLIFPPKPKDILRITRTPEVTCVVLDLGREDPEVQISWFVDGKEVHTAKTQSREQQFNGT
YRVVSVLPIEHQDWLTGKEFKCRVNHIDLPSPIERTISKARGRAHKPSVYVLPPSPKELSSSDTV
SITCLIKDFYPPDIDVEWQSNGQQEPERKHRMTPPQLDEDGSYFLYSKLSVDKSRWQQGDPFTCA
VMHETLQNHYTDLSLSHSPGK

cIgG-B [SEQ ID NO: 110]

LGGPSVFIFPPKPKDTLLIARTPEVTCVVVDLDPEDPEVQISWFVDGKQMQTAKTQPREEQFNGT
YRVVSVLPIGHQDWLKGKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVS
LTCLIKDFFPPDIDVEWQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAV
MHEALHNHYTQESLSHSPGK

cIgG-C [SEQ ID NO: 117]

LGGPSVFIFPPKPKDILVTARTPTVTCVVVDLDPENPEVQISWFVDSKQVQTANTQPREEQSNGT
YRVVSVLPIGHQDWLSGKQFKCKVNNKALPSPIEEIISKTPGQAHQPNVYVLPPSRDEMSKNTVT
LTCLVKDFFPPEIDVEWQSNGQQEPESKYRMTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAV
MHEALHNHYTQISLSHSPGK

cIgG-D [SEQ ID NO: 118]
LGGPSVFIFPPKPKDILRITRTPEITCVVLDLGREDPEVQISWFVDGKEVHTAKTQPREQQFNST
YRVVSVLPIEHQDWLTGKEFKCRVNHIGLPSPIERTISKARGQAHQPSVYVLPPSPKELSSSDTV
TLTCLIKDFFPPEIDVEWQSNGQPEPESKYHTTAPQLDEDGSYFLYSKLSVDKSRWQQGDTFTCA
VMHEALQNHYTDLSLSHSPGK

An amino acid sequence of the *kappa* canine light chain constant region is: [SEQ ID NO: 127]
RNDAQPAVYLFQPSPDQLHTGSASVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTY
SLSSTLTMSSTEYLSHELYSCEITHKSLPSTLIKSFQRSECQRVD

In the present invention, the amino acid sequence for each of the four canine IgG Fc fragments is based on the identified boundary of CH1 and CH2 domains as determined by Tang *et al, supra.* Caninized mouse anti-canine antibodies that bind canine IL-31RA include, but are not limited to: antibodies of the present invention that comprise canine IgG-A, IgG-B, IgG-C, and IgG-D heavy chains and/or canine *kappa* or *lambda* light chains together with mouse anti-canine IL-31RA CDRs. Accordingly, the present invention provides caninized mouse anti-canine antibodies of the present invention, including isolated caninized mouse anti-canine antibodies, that bind to canine IL-31RA and that preferably also block the binding of that canine IL-31RA to canine IL-31.

Accordingly, the present invention further provides caninized mouse antibodies and methods of use of the antibodies of the present invention in the treatment of a condition *e.g.*, the treatment of atopic dermatitis in canines.

The present invention further provides full length caninized heavy chains that can be matched with corresponding light chains to make a caninized antibody. Accordingly, the present invention further provides caninized mouse anti-canine antigen antibodies (including isolated caninized mouse anti-canine antibodies) of the present invention and methods of use of the antibodies of the present invention in the treatment of a condition *e.g.*, the treatment of atopic dermatitis in canines.

The present invention also provides antibodies of the present invention that comprise a canine fragment crystallizable region (cFc region) in which the cFc region has been genetically modified to augment, decrease, or eliminate one or more effector functions. In one aspect of the present invention, the genetically modified cFc region decreases or eliminates one or more effector functions. In another aspect of the invention the genetically modified cFc region

augments one or more effector function. In certain embodiments, the genetically modified cFc region is a genetically modified canine IgGB Fc region. In another such embodiment, the genetically modified cFc region is a genetically modified canine IgGC Fc region. In a particular embodiment the effector function is antibody-dependent cytotoxicity (ADCC) that is augmented, decreased, or eliminated. In another embodiment the effector function is complement-dependent cytotoxicity (CDC) that is augmented, decreased, or eliminated. In yet another embodiment, the cFc region has been genetically modified to augment, decrease, or eliminate both the ADCC and the CDC.

In order to generate variants of canine IgG that lack effector functions, a number of mutant canine IgGB heavy chains were generated. These variants may include one or more of the following single or combined substitutions in the Fc portion of the heavy chain amino acid sequence: P4A, D31A, N63A, G64P, T65A, A93G, and P95A. Variant heavy chains (*i.e.*, containing such amino acid substitutions) are cloned into expression plasmids and are transfected into HEK 293 cells along with a plasmid containing the gene encoding a light chain. Intact antibodies are expressed and purified from HEK 293 cells and then can be evaluated for binding to $Fc_{\gamma}RI$ and C1q to assess their potential for mediation of immune effector functions. [S*ee*, U.S. 10,106,607 B2, the contents of which are hereby incorporated by reference in its entirety.]

The present invention also provides modified canine IgG-Ds which in place of its natural IgG-D hinge region they comprise a hinge region from:

IgG-A:	FNECRCTDTPPCPVPEP	SEQ ID NO: 112
IgG-B:	PKRENGRVPRPPDCPKCPAPEM	SEQ ID NO: 113; or
IgG-C:	AKECECKCNCNNCPCPGCGL	SEQ ID NO: 114.

Alternatively, the IgG-D hinge region can be genetically modified by replacing a serine residue with a proline residue, *i.e.*, PKESTCKCI**P**PCPVPES, SEQ ID NO: 115 (with the proline residue (P) underlined and in bold substituting for the naturally occurring serine residue). Such modifications can lead to a canine IgG-D lacking fab arm exchange. The modified canine IgG-Ds can be constructed using standard methods of recombinant DNA technology [*e.g.*, Maniatis *et al.*, *Molecular Cloning, A Laboratory Manual* (1982)]. In order to construct these variants, the nucleic acids encoding the amino acid sequence of canine IgG-D can be modified so that it encodes the modified IgG-Ds. The modified nucleic acid sequences are then cloned into expression plasmids for protein expression.

The six complementary determining regions (CDRs) of a caninized mouse anti-canine antibody, as described herein can comprise a canine antibody *kappa* (*k*) or *lambda* (*l*) light chain

comprising a mouse light chain LCDR1, LCDR2, and LCDR3 and a canine antibody heavy chain IgG comprising a mouse heavy chain HCDR1, HCDR2, and HCDR3.

NUCLEIC ACIDS

The present invention further comprises the nucleic acids encoding the antibodies of the present invention (*see e.g.*, Examples below).

Also included in the present invention are nucleic acids that encode immunoglobulin polypeptides comprising amino acid sequences that are at least about 70% identical, preferably at least about 80% identical, more preferably at least about 90% identical and most preferably at least about 95% identical (*e.g.*, 95%, 96%, 97%, 98%, 99%, 100%) to the amino acid sequences of the caninized antibodies, with the exception of the CDRs which do not change, provided herein when the comparison is performed by a BLAST algorithm wherein the parameters of the algorithm are selected to give the largest match between the respective sequences over the entire length of the respective reference sequences. The present invention further provides nucleic acids that encode immunoglobulin polypeptides comprising amino acid sequences that are at least about 70% similar, preferably at least about 80% similar, more preferably at least about 90% similar and most preferably at least about 95% similar (*e.g.*, 95%, 96%, 97%, 98%, 99%, 100%) to any of the reference amino acid sequences when the comparison is performed with a BLAST algorithm, wherein the parameters of the algorithm are selected to give the largest match between the respective sequences over the entire length of the respective reference sequences, are also included in the present invention.

As used herein, nucleotide and amino acid sequence percent identity can be determined using C, MacVector (MacVector, Inc. Cary, NC 27519), Vector NTI (Informax, Inc. MD), Oxford Molecular Group PLC (1996) and the Clustal W algorithm with the alignment default parameters, and default parameters for identity. These commercially available programs can also be used to determine sequence similarity using the same or analogous default parameters. Alternatively, an Advanced Blast search under the default filter conditions can be used, *e.g.*, using the GCG (Genetics Computer Group, Program Manual for the GCG Package, Version 7, Madison, Wisconsin) pileup program using the default parameters.

The following references relate to BLAST algorithms often used for sequence analysis: BLAST ALGORITHMS: Altschul, S.F., *et al.*, *J. Mol. Biol.* 215:403-410 (1990); Gish, W., *et al.*, *Nature Genet.* 3:266-272 (1993); Madden, T.L., *et al.*, *Meth. Enzymol.* 266:131-141(1996); Altschul, S.F., *et al.*, *Nucleic Acids Res.* 25:3389-3402 (1997); Zhang, J., *et al.*, *Genome Res.* 7:649-656 (1997); Wootton, J.C., *et al.*, *Comput. Chem.* 17:149-163 (1993); Hancock, J.M. *et al.*,

Comput. Appl. Biosci. 10:67-70 (1994); ALIGNMENT SCORING SYSTEMS: Dayhoff, M.O., *et al.*, "A model of evolutionary change in proteins." in *Atlas of Protein Sequence and Structure*, vol. 5, suppl. 3. M.O. Dayhoff (ed.), pp. 345-352, (1978); *Natl. Biomed. Res. Found.*, Washington, DC; Schwartz, R.M., *et al.*, "Matrices for detecting distant relationships." in Atlas of Protein Sequence and Structure, vol. 5, suppl. 3." (1978), M.O. Dayhoff (ed.), pp. 353-358 (1978), *Natl. Biomed. Res. Found.*, Washington, DC; Altschul, S.F., *J. Mol. Biol.* 219:555-565 (1991); States, D.J., *et al.*, *Methods* 3:66-70(1991); Henikoff, S., *et al.*, *Proc. Natl. Acad. Sci.* USA 89:10915-10919 (1992); Altschul, S.F., *et al.*, *J. Mol. Evol.* 36:290-300 (1993); ALIGNMENT STATISTICS: Karlin, S., *et al.*, *Proc. Natl. Acad. Sci.* USA 87:2264-2268 (1990); Karlin, S., *et al.*, *Proc. Natl. Acad. Sci.* USA 90:5873-5877 (1993); Dembo, A., *et al.*, *Ann. Prob.* 22:2022-2039 (1994); and Altschul, S.F. "Evaluating the statistical significance of multiple distinct local alignments." in *Theoretical and Computational Methods in Genome Research* (S. Suhai, ed.), pp. 1-14, Plenum, New York (1997).

Antibody Protein Engineering

By way of example, and not limitation, the canine heavy chain constant region can be from IgGA, IgG-B, IgGC, IgGD, or a modified cFc, such as the IgG-Bm used herein [*see*, U.S. 10,106,607 B2, hereby incorporated by reference in its entirety] and the canine light chain constant region can be from *kappa* or *lambda*.

The antibodies can be engineered to include modifications to the canine framework and/or the canine frame residues within the variable domains of a parental (*i.e.*, mouse) monoclonal antibody, *e.g.* to improve the properties of the antibody.

The construction of caninized anti-canine IL-31 receptor *alpha* monoclonal antibodies can be performed by determining a DNA sequence that encodes the heavy and light chains of canine IgG were determined. The DNA and protein sequence of the canine heavy and light chains are known in the art and can be obtained by searching of the NCBI gene and protein databases. As indicated above, for canine antibodies there are four known IgG subtypes: IgG-A, IgG-B, IgG-C, and IgG-D, and two types of light chains, *i.e.*, *kappa* and *lambda*.

A caninized mouse anti-canine IL-31RA antibody can be produced recombinantly by methods that are known in the field. Mammalian cell lines available as hosts for expression of the antibodies or fragments disclosed herein are well known in the art and include many immortalized cell lines available from the American Type Culture Collection (ATCC). These include, *inter alia*, Chinese hamster ovary (CHO) cells, NSO, SP2 cells, HeLa cells, baby hamster kidney (BHK) cells, monkey kidney cells (COS), human hepatocellular carcinoma cells

(*e.g.*, Hep G2), A549 cells, 3T3 cells, HEK-293 cells and a number of other cell lines. Mammalian host cells include human, mouse, rat, dog, monkey, pig, goat, bovine, horse and hamster cells. Cell lines of particular preference are selected through determining which cell lines have high expression levels. Other cell lines that may be used are insect cell lines, such as Sf9 cells, amphibian cells, bacterial cells, plant cells and fungal cells. When recombinant expression vectors encoding the heavy chain or antigen-binding portion or fragment thereof, the light chain and/or antigen-binding fragment thereof are introduced into mammalian host cells, the antibodies are produced by culturing the host cells for a period of time sufficient to allow for expression of the antibody in the host cells or, more preferably, secretion of the antibody into the culture medium in which the host cells are grown.

Antibodies can be recovered from the culture medium using standard protein purification methods. Further, expression of antibodies of the invention (or other moieties therefrom) from production cell lines can be enhanced using a number of known techniques. For example, the glutamine synthetase gene expression system (the GS system) is a common approach for enhancing expression under certain conditions. The GS system is discussed in whole or part in connection with European Patent Nos. 0 216 846, 0 256 055, and 0 323 997 and European Patent Application No. 89303964.4.

In certain embodiments, the antibody or antigen binding fragment comprises a heavy chain constant region, *e.g.*, a canine constant region, such as IgG-A, IgG-B, IgG-C and IgG-D canine heavy chain constant region or a variant thereof. In certain embodiments, the antibody or antigen binding fragment comprises a light chain constant region, *e.g.*, a canine light chain constant region, such as *lambda* or *kappa* canine light chain region or variant thereof. By way of example, and not limitation, the canine heavy chain constant region can be from IgG-B and the canine light chain constant region can be from *kappa*.

EPITOPE MAPPING

The interaction of antibodies with their cognate protein antigens is mediated through the binding of specific amino acids of the antibodies (paratopes) with specific amino acids (epitopes) of target antigens. An epitope is an antigenic determinant that causes a specific reaction by an immunoglobulin. An epitope consists of a group of amino acids on the surface of the antigen. A protein of interest may contain several epitopes that are recognized by different antibodies. The epitopes recognized by antibodies are classified as linear or conformational epitopes. Linear epitopes are formed by a stretch of a continuous sequence of amino acids in a protein, while

conformational epitopes are composed of amino acids that are discontinuous (*e.g.*, far apart) in the primary amino acid sequence, but are brought together upon three-dimensional protein folding.

Epitope mapping refers to the process of identifying the amino acid sequences (*i.e.*, epitopes) that are recognized by antibodies on their target antigens. Identification of epitopes recognized by monoclonal antibodies (mAbs) on target antigens has important applications. For example, it can aid in the development of new therapeutics, diagnostics, and vaccines. Epitope mapping can also aid in the selection of optimized therapeutic mAbs and help elucidate their mechanisms of action. Epitope information on IL-31 receptor *alpha* can also elucidate unique epitopes and define the protective or pathogenic effects of vaccines. Epitope identification also can lead to development of subunit vaccines based on chemical or genetic coupling of the identified peptide epitope to a carrier protein or other immunostimulating agents.

Epitope mapping can be carried out using polyclonal or monoclonal antibodies and several methods are employed for epitope identification depending on the suspected nature of the epitope (*i.e.*, linear *versus* conformational). Mapping linear epitopes is more straightforward and relatively, easier to perform. For this purpose, commercial services for linear epitope mapping often employ peptide scanning. In this case, an overlapping set of short peptide sequences of the target protein are chemically synthesized and tested for their ability to bind antibodies of interest. The strategy is rapid, high-throughput, and relatively inexpensive to perform. On the other hand, mapping of a discontinuous epitope is more technically challenging and requires more specialized techniques such as x-ray co-crystallography of a monoclonal antibody together with its target protein, Hydrogen-Deuterium (H/D) exchange, Mass Spectrometry coupled with enzymatic digestion as well as several other methods known to those skilled in the art.

Epitope Binding and Cross-Blocking Antibodies

An anti-canine IL-31RA antibody or antigen-binding fragment thereof of the present invention includes any antibody or antigen-binding fragment thereof that binds to the same epitope in canine IL-31RA as the one of the antibodies, disclosed herein, bind, *e.g.*, such as the 218D9 antibody which binds to the epitope comprising the amino acid sequence either SEQ ID NO: 104, SEQ ID NO: 105, or both SEQ ID NO: 104 and SEQ ID NO: 105, or the 51F8 antibody which binds to the epitope comprising the amino acid sequence either SEQ ID NO: 98, SEQ ID NO: 100, or both SEQ ID NO: 98 and SEQ ID NO: 100, including caninized antibodies, and any antibody or antigen-binding fragment that cross-blocks (partially or fully) or is cross-

blocked (partially or fully) by an antibody or fragment discussed herein for canine IL-31RA binding; as well as any variant thereof.

The cross-blocking antibodies and antigen-binding fragments can be identified based on their ability to cross-compete with *e.g.*, the 100H8 or 74H10 antibody in standard binding assays (*e.g.*, BIACore®, ELISA, as exemplified below, or flow cytometry). For example, standard ELISA assays can be used in which a recombinant canine IL-31RA protein is immobilized on the plate, one of the antibodies is fluorescently labeled and the ability of non-labeled antibodies to compete off the binding of the labeled antibody is evaluated. Additionally or alternatively, BIAcore® analysis can be used to assess the ability of the antibodies to cross-compete. The ability of a test antibody to inhibit the binding of the *e.g.*, 100H8 or 74H10 antibody, to canine IL-31RA demonstrates that the test antibody can compete with the 100H8 or 74H10 antibody for binding to canine IL-31RA and thus, may, in some cases, bind to the same epitope on canine IL-31RA as the 100H8 and/or 74H10 antibody binds.

Antibodies and fragments thereof that bind to the same epitope as any of the anti-canine IL-31RA antibodies or fragments of the present invention also form part of the present invention.

PHARMACEUTICAL COMPOSITIONS AND ADMINISTRATION

To prepare pharmaceutical or sterile compositions comprising the antibodies of the present invention, these antibodies can be admixed with a pharmaceutically acceptable carrier or excipient. [*See, e.g.*, *Remington's Pharmaceutical Sciences* and *U.S. Pharmacopeia: National Formulary*, Mack Publishing Company, Easton, PA (1984)].

Formulations of therapeutic and diagnostic agents may be prepared by mixing with acceptable carriers, excipients, or stabilizers in the form of, *e.g.*, lyophilized powders, slurries, aqueous solutions or suspensions [see, *e.g.*, Hardman, *et al.* (2001) *Goodman and Gilman's The Pharmacological Basis of Therapeutics*, McGraw-Hill, New York, NY; Gennaro (2000) *Remington: The Science and Practice of Pharmacy*, Lippincott, Williams, and Wilkins, New York, NY; Avis, *et al.* (eds.) (1993) *Pharmaceutical Dosage Forms: Parenteral Medications*, Marcel Dekker, NY; Lieberman, *et al.* (eds.) (1990) *Pharmaceutical Dosage Forms: Tablets*, Marcel Dekker, NY; Lieberman, *et al.* (eds.) (1990) *Pharmaceutical Dosage Forms: Disperse Systems*, Marcel Dekker, NY; Weiner and Kotkoskie (2000) *Excipient Toxicity and Safety*, Marcel Dekker, Inc., New York, NY]. In one embodiment, the antibodies of the present invention are diluted to an appropriate concentration in a sodium acetate solution pH 5-6, and

NaCl or sucrose is added for tonicity. Additional agents, such as polysorbate 20 or polysorbate 80, may be added to enhance stability.

Toxicity and therapeutic efficacy of the antibody compositions, administered alone or in combination with another agent, can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, *e.g.*, for determining the LD_{50} (the dose lethal to 50% of the population) and the ED_{50} (the dose therapeutically effective in 50% of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index (LD_{50}/ ED_{50}). In particular aspects, antibodies exhibiting high therapeutic indices are desirable. The data obtained from these cell culture assays and animal studies can be used in formulating a range of dosage for use in canines. The dosage of such compounds lies preferably within a range of circulating concentrations that include the ED_{50} with little or no toxicity. The dosage may vary within this range depending upon the dosage form employed and the route of administration.

The mode of administration can vary. Suitable routes of administration include oral, rectal, transmucosal, intestinal, parenteral; intramuscular, subcutaneous, intradermal, intramedullary, intrathecal, direct intraventricular, intravenous, intraperitoneal, intranasal, intraocular, inhalation, insufflation, topical, cutaneous, transdermal, or intra-arterial. In particular embodiments, the antibodies of the present invention can be administered by an invasive route such as by injection. In further embodiments of the invention, the antibodies of the present invention, or pharmaceutical composition thereof, is administered intravenously, subcutaneously, intramuscularly, intraarterially, or by inhalation, aerosol delivery. Administration by non-invasive routes (*e.g.*, orally; for example, in a pill, capsule or tablet) is also within the scope of the present invention.

Compositions can be administered with medical devices known in the art. For example, a pharmaceutical composition of the invention can be administered by injection with a hypodermic needle, including, *e.g.*, a prefilled syringe or autoinjector. The pharmaceutical compositions disclosed herein may also be administered with a needleless hypodermic injection device; such as the devices disclosed in U.S. Patent Nos.: 6,620,135; 6,096,002; 5,399,163; 5,383,851; 5,312,335; 5,064,413; 4,941,880; 4,790,824 or 4,596,556.

The pharmaceutical compositions disclosed herein may also be administered by infusion. Examples of well-known implants and modules form administering pharmaceutical compositions include: U.S. Patent No. 4,487,603, which discloses an implantable micro-infusion pump for dispensing medication at a controlled rate; U.S. Patent No. 4,447,233, which discloses a medication infusion pump for delivering medication at a precise infusion rate; U.S. Patent No.

4,447,224, which discloses a variable flow implantable infusion apparatus for continuous drug delivery; U.S. Patent. No. 4,439,196, which discloses an osmotic drug delivery system having multi-chamber compartments. Many other such implants, delivery systems, and modules are well known to those skilled in the art.

Alternatively, one may administer the antibodies of the present invention in a local rather than systemic manner, often in a depot or sustained release formulation.

The administration regimen depends on several factors, including the serum or tissue turnover rate of the therapeutic antibodies, the level of symptoms, the immunogenicity of the therapeutic antibodies and the accessibility of the target cells in the biological matrix. Preferably, the administration regimen delivers sufficient therapeutic antibodies to effect improvement in the target disease/condition state, while simultaneously minimizing undesired side effects. Accordingly, the amount of biologic delivered depends in part on the particular therapeutic antibodies and the severity of the condition being treated. Guidance in selecting appropriate doses of therapeutic antibodies is available [*see, e.g.*, Wawrzynczak *Antibody Therapy*, Bios Scientific Pub. Ltd, Oxfordshire, UK (1996); Kresina (ed.) *Monoclonal Antibodies, Cytokines and Arthritis*, Marcel Dekker, New York, NY (1991); Bach (ed.) *Monoclonal Antibodies and Peptide Therapy in Autoimmune Diseases*, Marcel Dekker, New York, NY (1993); Baert, *et al. New Engl. J. Med.* 348:601-608 (2003); Milgrom *et al. New Engl. J. Med.* 341:1966-1973 (1999); Slamon *et al. New Engl. J. Med.* 344:783-792 (2001); Beniaminovitz *et al. New Engl. J. Med.* 342:613-619 (2000); Ghosh *et al. New Engl. J. Med.* 348:24-32 (2003); Lipsky *et al. New Engl. J. Med.* 343:1594-1602 (2000)].

Determination of the appropriate dose is made by the veterinarian, *e.g.*, using parameters or factors known or suspected in the art to affect treatment. Generally, the dose begins with an amount somewhat less than the optimum dose and it is increased by small increments thereafter until the desired or optimum effect is achieved relative to any negative side effects. Important diagnostic measures include those of the symptoms.

Antibodies provided herein may be provided by continuous infusion, or by doses administered, *e.g.*, daily, 1-7 times per week, weekly, bi-weekly, monthly, bimonthly, quarterly, semiannually, annually etc. Doses may be provided, *e.g.*, intravenously, subcutaneously, topically, orally, nasally, rectally, intramuscular, intracerebrally, intraspinally, or by inhalation. A total weekly dose is generally at least 0.05 μg/kg body weight, more generally at least 0.2 μg/kg, 0.5 μg/kg, 1 μg/kg, 10 μg/kg, 100 μg/kg, 0.25 mg/kg, 1.0 mg/kg, 2.0 mg/kg, 5.0 mg/ml, 10 mg/kg, 25 mg/kg, 50 mg/kg or more [*see, e.g.*, Yang, *et al. New Engl. J. Med.* 349:427-434

(2003); Herold, *et al. New Engl. J. Med.* 346:1692-1698 (2002); Liu, *et al. J. Neurol. Neurosurg. Psych.* 67:451-456 (1999); Portielji, *et al. Cancer Immunol. Immunother.* 52:133-144 (2003)]. Doses may also be provided to achieve a pre-determined target concentration of antibodies of the present invention in the canine's serum, such as 0.1, 0.3, 1, 3, 10, 30, 100, 300 μg/ml or more. In other embodiments, antibodies of the present invention is administered subcutaneously or intravenously, on a weekly, biweekly, "every 4 weeks," monthly, bimonthly, or quarterly basis at 10, 20, 50, 80, 100, 200, 500, 1000 or 2500 mg/subject.

As used herein, "inhibit" or "treat" or "treatment" includes a postponement of development of the symptoms associated with a disorder and/or a reduction in the severity of the symptoms of such disorder. The terms further include ameliorating existing uncontrolled or unwanted symptoms, preventing additional symptoms, and ameliorating or preventing the underlying causes of such symptoms. Thus, the terms denote that a beneficial result has been conferred on a vertebrate subject (*e.g.*, a canine) with a disorder, condition and/or symptom, or with the potential to develop such a disorder, disease or symptom.

As used herein, the terms "therapeutically effective amount", "therapeutically effective dose" and "effective amount" refer to an amount of antibodies of the present invention that, when administered alone or in combination with an additional therapeutic agent to a cell, tissue, or subject, *e.g.*, canine, is effective to cause a measurable improvement in one or more symptoms of a disease or condition or the progression of such disease or condition. A therapeutically effective dose further refers to that amount of the antibodies sufficient to result in at least partial amelioration of symptoms, *e.g.*, treatment, healing, prevention or amelioration of the relevant medical condition, or an increase in rate of treatment, healing, prevention or amelioration of such conditions. When applied to a combination, a therapeutically effective dose refers to combined amounts of the active ingredients that result in the therapeutic effect, whether administered in combination, serially, or simultaneously. An effective amount of a therapeutic will result in an improvement of a diagnostic measure or parameter by at least 10%; usually by at least 20%; preferably at least about 30%; more preferably at least 40%, and most preferably by at least 50%. An effective amount can also result in an improvement in a subjective measure in cases where subjective measures are used to assess severity of the condition.

EXAMPLES

EXAMPLE 1

IL-31 RECEPTOR *alpha*

Nucleotide Sequence

The nucleotide sequence of SEQ ID NO: 1 encodes the extracellular domain of the canine IL-31 receptor *alpha* (cIL-31RA) fused to a HIS tag. Canine IL-31RA ECD HIS-tagged protein comprises the amino acid sequence of SEQ ID NO: 2. The nucleotide sequence was prepared by chemical synthesis and then cloned into expression plasmids that are suitable for production of the corresponding proteins in eukaryotic cells, either HEK-293 or CHO cells.

Canine IL-31RA ECD-10His: [SEQ ID NO: 1]

gtgctgcccgccaagcccgagaacatcagctgcatcttctactacgaggagaacttcacctgcacctggag
ccccgagaaggaggccagctacacctggtacaaggtgaagagaacctacagctacggctacaagagcgaca
tctgcagcaccgacaacagcaccagaggcaaccacgccagctgcagcttcctgccccccaccatcaccaac
cccgacaactacaccatccaggtggaggcccagaacgccgacggcatcatgaagagcgacatcacctactg
gaacctggacgccatcatgaagatcgagccccccgagatcttcagcgtgaagagcgtgctgggcatcaaga
gaatgctgcagatcaagtggatcagacccgtgctggccccccacagcagcaccctgaagtacaccctgaga
ttcagaaccatcaacagcgcctactggatggaggtgaacttcaccaaggaggacatcgacagagacgagac
ctacaacctgaccgagctgcaggccttcaccgagtacgtgatgaccctgagatgcgccccgccgagagca
tgttctggagcggctggagccaggagaaggtgggcaccaccgaggaggaggccccctacggcctggacctg
tggagagtgctgaagcccgccatggtggacggcagaagacccgtgcagctgatgtggaagaaggccaccgg
cgcccccgtgctggagaaggccctgggctacaacatctggtacttccccgagaacaacaccaacctgaccg
agaccgtgaacaccaccaaccagacccacgagctgtacctgggcggcaagacctactgggtgtacgtggtg
agctacaacagcctgggcgagagccccgtggccaccctgagaatccccgccctgaacgagaagaccttcca
gtgcatcgaggccatgcaggcctgcctgacccaggaccagctggtggtggagtggcagagcagcgccccg
aggtggacacctggatggtggagtggttccccgacgtggacagcgagcccagcagcttcagctgggagagc
gtgagccaggccagaaactggaccatccagaaggacgagctgaagcccctgtggtgctacaacatcagcgt
gtaccccgtgctgagagacagagtgggccagccctacagcacccaggcctacgtgcaggagggcatcccca
gcgccggccccgtgacccaggccgacagcatcggcgtgaagaccgtgaccatcacctggaaggagatcccc
aagagcaagagaaacggcttcatcaagaactacaccatcttctaccaggccgaggacggcaaggagttcag
caagaccgtgaacagcaacatcctgcagtacagactggagagcctgaccagaagaaccagctacagcctgc
aggtgatggccagcaccaacgccggcggcaccaacggcaccaagatcaacttcaagaccctgagcatcagc
caccaccaccaccaccaccaccaccac

EXAMPLE 2

EXPRESSION AND PURIFICATION OF IL-31 RECEPTOR *alpha* ECD

Plasmids comprising the nucleotide sequence of SEQ ID NO: 1 were transfected into HEK-293 or CHO cells using electroporation *via* the MaxCyte instrument as per the manufacturer's recommendation. Several days following transfection, the supernatants of transfected cells and un-transfected controls were harvested and spun down to remove cellular debris. IL-31RA with the HIS tag was purified from cell culture fluids by passing the clarified harvested fluid from transfected cells over nickel columns as per the manufacturer's recommendation. Purified proteins were quantified by measuring their absorbance of ultraviolet light at 280 nm.

Canine IL-31RA ECD-10His: [SEQ ID NO: 2]

VLPAKPENISCIFYYEENFTCTWSPEKEASYTWYKVKRTYSYGYKSDICSTDNSTRGNHASCSFL
PPTITNPDNYTIQVEAQNADGIMKSDITYWNLDAIMKIEPPEIFSVKSVLGIKRMLQIKWIRPVL
APHSSTLKYTLRFRTINSAYWMEVNFTKEDIDRDETYNLTELQAFTEYVMTLRCAPAESMFWSGW
SQEKVGTTEEEAPYGLDLWRVLKPAMVDGRRPVQLMWKKATGAPVLEKALGYNIWYFPENNTNLT
ETVNTTNQTHELYLGGKTYWVYVVSYNSLGESPVATLRIPALNEKTFQCIEAMQACLTQDQLVVE
WQSSAPEVDTWMVEWFPDVDSEPSSFSWESVSQARNWTIQKDELKPLWCYNISVYPVLRDRVGQP
YSTQAYVQEGIPSAGPVTQADSIGVKTVTITWKEIPKSKRNGFIKNYTIFYQAEDGKEFSKTVNS
NILQYRLESLTRRTSYSLQVMASTNAGGTNGTKINFKTLSISHHHHHHHHHH

TABLE 1

CANINE IL-31RA EXTRACELLULAR DOMAIN -HIS TAG

	SEQ ID NO:	Nucleic Acid	Amino Acid
canine IL-31RA ECD-10His	1	X	
canine IL-31RA ECD-10His	2		X

EXAMPLE 3

BINDING OF CANINE IL-31RA TO BIOTINYLATED CANINE IL-31:

Protocol

1) Coat immunoplate(s) with IL-31RA proteins by diluting to 1 µg/mL in phosphate-buffered saline solution (PBS). Add 100µL/well. Incubate the plate(s) at 2-7° overnight.

2) Wash the plates 3 times with 275 µL/well of phosphate-buffered saline solution plus TWEEN 20 (PBST).

3) Block the plates with 200 µL/well of blocking buffer [1% nonfat dried milk (NFDM) in PBST] for 30-45 minutes at 36 ± 2°C with gentle shaking (120 ± 20 RPM).

4) Wash the plates 3 times with 275 µL/well of PBST.

5) 3-fold dilute biotinylated IL-31 (at 10 µg/mL) in 1% NFDM in PBST on a dilution plate, and transfer 100 µL/well to the immunoplate(s). Incubate for 30-45 minutes at 36 ± 2°C with gentle shaking (120 ± 20 RPM).

6) Wash the plates 3 times with 275 µL/well of PBST.

7) Dilute horse raddish peroxidase-Streptavidin (HRP-Streptavidin) to a final dilution of 1:1000 in 1% NFDM in PBST.

8) Add 100 µL/well of HRP-Streptavidin to the immunoplate(s) and incubate for 30-45 minutes at 36 ± 2°C with gentle shaking (120 ± 20 RPM).

9) Wash the plates 3 times with 275 µL/well of PBST.

10) Combine equal volumes of pre-warmed TMP 2-Component substrate immediately before use.

11) Add 100 µL/well of prepared 3,3',5,5'-tetramethylbenzidine (TMP) substrate to the immunoplate(s) and incubate in the dark for 10 to 15 minutes at 36 ± 2°C with gentle shaking (120 ± 20 RPM).

12) Stop the reaction by addition of 100 µL/well of 1 M H3PO4.

13) Read the plates using a microplate reader at a wavelength of 450 nm with a reference wavelength of 540 nm.

The extracellular domain (ECD) of canine IL-31RA was tested for its ability to bind to canine IL-31 (*see,* Figure 1). The results indicate that IL-31RA ECD binds in a dose-dependent manner to biotinylated canine IL-31 with an EC50 of 0.55 ng/ml.

EXAMPLE 4

MONOCLONAL ANTIBODIES AGAINST CANINE IL-31 RECEPTOR *alpha*

Monoclonal antibodies (mAbs) against canine IL-31RA were produced by the immunization of mice multiple times with canine IL-31RA ECD. Mice were immunized *via* the Intraperitoneal route with IL-31RA ECD in GS proprietary adjuvant 3 times on days 0, 14, and 28 using 50 µg per mouse for first immunization and 25 µg per mouse for the subsequent boosts. On day 48 mice were immunized once more with 25 µg of antigen and 4 days later their spleen cells were fused with the myeloma SP2/0 cell line to produce hybridomas secreting antibodies. At various time points after immunization, sera were collected from mice and tested against canine IL-31RA by ELISA. The spleen cells from the mouse with highest IL-31RA ECD reactivity were fused with the myeloma SP2/0 cell line to produce hybridomas. Approximately

14 days after the fusion, supernatants from growing hybridomas were screened by flow cytometry using cells expressing the IL-31RA protein. The reactivities of hybridoma were confirmed by ELISA as follows:

Procedure for the ELISA:

1) Coat 96-well plates with IL-31RA (1 μg/mL in PBS buffer), 25 μL/well.

2) Incubate the plates at 4°C overnight.

3) Wash the plates 3 times with PBST (PBS +0.05% Tween 20)

4) Block the plates with blocking buffer [PBS with 5% fetal bovine serum (FBS)], 25μl/well for 30 minutes at room temperature.

5) Transfer 25 μl/well hybridoma supernatant to the 96-well plates, incubate 60 minutes at room temperature.

6) Wash the plates 3 times by PBST.

7) Add 25μl/well anti-mouse HRP, 1:4000 dilution in blocking buffer, to the plates and incubate 60 minutes at room temperature.

8) Wash the plates 5 times by PBST.

9) Add TMB based reagent to the plates for colorimetric reaction for 2-3 minutes.

10) Stop the reactions with 0.16M sulfuric acid.

11) Read the plates by plate reader.

As shown in Figures 2A-2B, the selected mouse mAbs were tested for their reactivity to canine IL-31RA. The results indicate that the selected mouse mAbs bind to canine IL-31RA in a dose-dependent manner. Ten (10) mouse monoclonal antibodies were obtained that have strong binding reactivity to canine IL-31RA, as shown in Figures 2A-2B.

The amino acid sequences of the heavy and light chain variable regions of these ten mouse monoclonal antibody are provided below.

100H8VH [SEQ ID NO: 52]

EVQLQQSGPELVKPGASVKIPCKASGYTFTDYNMDWVKQSHGKSLEWIGHINPNNGGILYNQKFK
GKATLTVDRSSNTAYMELRSLTSEDTAVYFCARWAPLVRQPYWYFDVWGTGTTVTVSS

100H8VL [SEQ ID NO: 53]

DIKMTQSPSSIFASLGERVTITCKASQDINSYLNWFQQKPGKSPKTLIYRADRLVDGVPSRFSGS
GSGQDYSLTISSLEYEDMGIYYCLQYDEFPLTFGAGTKLELN

222E7VH [SEQ ID NO: 54]

EVQLQQSGPELVKPGSSVKIPCKASGYTFTDYNMDWVKQSHGKSLEWIGHINPNIGGTLYNQKFK
GKATLSVDKSSSTAYMELRSLTSEDTAVYYCARWAQLQRQPYWYFDVWGPGTTVTVSS

222E7VL [SEQ ID NO: 55]
DIKMTQSPSSMYASLGERVTITCKASQDINSYLNWFQQKPGKSPKTLIYRVNRLVDGVPSRFSGS
GSGQDYSLTISSLEYEDMGIYYCLQYDEFPLTFGAGTKLELK

55B3VH [SEQ ID NO: 56]
EVQLQQSGPELVKPGASVKIPCKASGYTFTDYNMDWVKQSHGKSLEWIGHINPNTGGTIYNQKFK
GKATLTVDKSSSTAYMDLRSLTSEDTAVYYCARWAQLLRQPYWYFDVWGTGTTVTVSS

55B3VL [SEQ ID NO: 57]
DIKMTQSPSSMYASLGERVTITCKASQDINSYFNWVQQKPGKSPKTLIFRANRLVDGVPSRFSGS
GSGQDYSLTINSLEYEDMGIYYCLQYDEFPLTFGAGTKLELK

74H10VH [SEQ ID NO: 58]
EVQLQQSGPELVKPGASMKIPCKTSGYTFTDYNMDWVKQSHGKSLEWIGHINPNNGGTLYNQKFK
DKATLTVDRSSNTAYMELRSLTSEDTAVYYCARWAPLLRQPYWYFDVWGTGTTVTVSS

74H10VL [SEQ ID NO: 59]
DIKMTQSPSSMYASLGERVTITCKASQDINSYLNWFQQKPGKSPKTLIYRADRLVDGVPSRFSGS
GSGQDYSLTISSLEYEDMGIYYCLQYDEFPLTFGDGTKLELN

209G5VH [SEQ ID NO: 60]
EVQLQQSVAELVRPGASVKLSCTASGFNIKNTYMHWVKERPEQGLEWIGRIDPANGNSKYAPKFQ
GKATITTDTSSNTAYLQLSSLTSEDTAIYYCARYYYVSSHFDVWGTGTTVTVSS

209G5VL [SEQ ID NO: 61]
SIVMTQTPKFLLVSAGDRVTITCKASQSVTNDVTWYQQKPGQSPKLLIYYASNRYT
GVPDRFTGSGYGTDFTFTISTVQAEDLAVYFCQQDYSSPFTFGSGTKLEIK

51F8VH [SEQ ID NO: 62]

EVQLQQSVAELVRPGASVKLSCTASGFNIKNTFIHWVKQRPEQGLEWIGRIDPANGNTKYAPKFQ
GRATLTADTSSNTAYLQLSSLTSEDTAIYYCARYYYVSSYFDVWGTGTTVTVSS

51F8VL [SEQ ID NO: 63]
SIVMTQTPKFLLVSAGDRVTITCKASQSVTNDVTWYQQKPGQSPKLLIFSASNRYTGVPDRFTGS
GYGTDFTFTISTVQAEDLAVYFCQQDYSSPWTFGGGTKLEIK

65G9VH [SEQ ID NO: 64]
EFQLQQSGPELVKPGASVKISCKASGYSFTDYGMNWLKQINGKSLEWIAIINPNYGTASSNPKFK
DKATLTVDHSSSTAYMQLSSLTSEDSAVYYCARAFDGYYFYWYFDVWGTGTTVTVSS

65G9VL [SEQ ID NO: 65]
DVVMTQTPLSLPVSLGDQASISCRSSQSLIHTNGNTYLHWYLQKPGQSPKLLIYKVSNRFSGVPD
RFSGSGSGTDFTLKISRLEAEDLGVYFCSQSTHVPLTFGAGTKLELR

85C10VH [SEQ ID NO: 66]
EFQLQQSGPELVKPGASVKISCKASGYSFTDYSINWVKQSNGKSLEWIGVINPNYGTSSHNQKFK
GKATMTVDQSSSTAYMQLNSLTSEDSAVYYCARALDDYYFYWYFDVWGIGTTVTVSS

85C10VL [SEQ ID NO: 67]
DVVMTQTPLSLPVSLGDQASISCRSSQSLVHTNGNTYLHWYLQKPGQSPKLLIYKVSNRFSGVPD
RFSGSGSGTDFTLKISRVEAEDLGVYFCSQTTHVPLTFGAGTKLELK

218D9VH [SEQ ID NO: 68]
QVTLKESGPGILQPSQTLSLTCSFSGFSLSTFGRGVGWIRQPSGKGLDWLTHIWWDDDKYYNPAL
KSRLTISKDTSKNQVFLKIANVDTADTATYYCARIAGGLRRAPYAMDSWGQGTSVTVSS

218D9VL [SEQ ID NO: 69]
DIQMTQSPASLSVSVGETVTITCRASENIYSSLAWYQQKQGKSPQLLVYAATNLADGVPSRFSGS
GSGTQYSLKINSLQSEDSGNYYCQHFRDTPPTFGGGTKLEIK

224G3VH [SEQ ID NO: 70]

EVQLVESGGDLVKPGGSLKLSCAASGFTFSSYGMSWVRQTPDKRLEWVATISNGGSYTYYPDSVK
GRFTISRDNAKNTLYMQMSSLKSEDTAMYYCARNEPPDYWGQGTSVTVSS

224G3VL [SEQ ID NO: 71]

QIVLTQSPAIMSASLGERVTMTCTASSSVGSSYLHWYQQKPGSSPKLWIYDTSNLASGVPVRFSG
SGSGTSYSLTISSMEAEDAATYYCHQYHRSPYTFGGGTKLEIK

Table 2 below, provides the association rate constant (ka), dissociation rate constant (kd), and dissociation constant (KD), as analyzed by Octet Kinetics (*see also*, Example 9 below). These constants reflect the binding affinity of the individual monoclonal antibodies for canine IL-31 RA.

TABLE 2

BINDING AFFINITY OF SELECTED ANTI-IL-31RA ANTIBODIES

mAb	KD (M)	ka (1/Ms)	kd (1/s)
100H8	1.52E-10	7.88E+05	1.20E-04
51F8	2.07E-10	6.84E+05	1.41E-04
55B3	2.72E-10	5.68E+05	1.54E-04
65G9	4.51E-10	7.87E+05	3.55E-04
74H10	1.96E-10	4.63E+05	9.05E-05
209G5	1.31E-10	3.70E+05	4.87E-05
85C10	8.51E-10	4.27E+05	3.63E-04
218D9	6.98E-11	6.64E+05	4.63E-05
222E7	1.74E-10	1.62E+06	2.82E-04
224G3	9.64E-13	1.12E+06	1.08E-06

The results show that the selected mAbs have low nanomolar to sub-picomolar binding affinities ranging from about 0.85 nM to about 1 pM.

The sets of the six CDRs for each of the ten monoclonal antibodies described above are provided below in Table 3. In addition, the canonical structures for each of these CDRs are provided in Table 4 below.

TABLE 3

AMINO ACID SEQUENCES OF THE MOUSE CDRS

	Amino Acid Sequence	SEQ ID NO:	Amino Acid Sequence	SEQ ID NO:
	100H8		74H10	
HCDR1	DYNMD	3	DYNMD	3
HCDR2	HINPNNGGILYNQKFKG	10	HINPNNGGTLYNQKFKD	11
HCDR3	WAPLVRQPYWY	20	WAPLLRQPYWYFDV	21
LCDR1	KASQDINSYLN	30	KASQDINSYLN	30
LCDR2	RADRLVD	40	RADRLVD	40
LCDR3	LQYDEFPLT	45	LQYDEFPLT	45
	222E7		55B3	
HCDR1	DYNMD	3	DYNMD	3
HCDR2	HINPNIGGTLYNQKFKG	12	HINPNTGGTIYNQKFKG	13
HCDR3	WAQLQRQPYWYFDV	22	WAQLLRQPYWYFDV	23
LCDR1	KASQDINSYLN	30	KASQDINSYFN	31
LCDR2	RVNRLVD	38	RANRLVD	39
LCDR3	LQYDEFPLT	45	LQYDEFPLT	45
	51F8		209G5	
HCDR1	NTFIH	5	NTYMH	6
HCDR2	RIDPANGNTKYAPKFQG	15	RIDPANGNSKYAPKFQG	16
HCDR3	YYYVSSY	25	YYYVSSHFDV	26
LCDR1	KASQSVTNDVT	32	KASQSVTNDVT	32
LCDR2	SASNRYT	41	YASNRYT	42
LCDR3	QQDYSSPWT	47	QQDYSSPFT	48
	65G9		218D9	
HCDR1	DYGMN	7	TFGRGVG	8
HCDR2	IINPNYGTASSNPKFKD	17	HIWWDDDKYYNPALKS	18
HCDR3	AFDGYYFYWYFDV	27	IAGGLRRAPYAMDS	28
LCDR1	RSSQSLIHTNGNTYLH	34	RASENIYSSLA	35
LCDR2	KVSNRFS	37	AATNLAD	43
LCDR3	SQSTHVPLT	50	QHFRDTPPT	51
	224G3		85C10	
HCDR1	SYGMS	9	DYSIN	4
HCDR2	TISNGGSYTYYPDSVKG	19	VINPNYGTSSHNQKFKG	14

HCDR3	NEPPDY	29	ALDDYYFYWY	24
LCDR1	TASSSVGSSYLH	36	RSSQSLVHTNGNTYLH	33
LCDR2	DTSNLAS	44	KVSNRFS	37
LCDR3	HQYHRSPYT	49	SQTTHVPLT	46

TABLE 4

CANONICAL STRUCTURE OF THE MOUSE CDRs

	L1	L2	L3	H1	H2	H3
100H8	L1-2	L2-1	L3-1	H1-1	H2-2	H3-11
85C10	L1-4	L2-1	L3-1	H1-1	H2-2	H3-10
51F8	L1-2	L2-1	L3-1	H1-1	H2-2	H3-7
222E7	L1-2	L2-1	L3-1	H1-1	H2-2	H3-14
55B3	L1-2	L2-1	L3-1	H1-1	H2-2	H3-14
209G5	L1-2	L2-1	L3-1	H1-1	H2-2	H3-10
65G9	L1-4	L2-1	L3-1	H1-1	H2-2	H3-14
218D9	L1-2	L2-1	L3-1	H1-1	H2-4	H3-14
224G3	L1-2	L2-1	L3-1	H1-1	H2-4	H3-6
74H10	L1-2	L2-1	L3-1	H1-1	H2-2	H3-14

Of these 10 sets of CDRS in Table 3 above, a particular group of four anti-canine IL-31RA antibodies that bind IL-31RA were identified as comprising a striking amino acid sequence similarity.

HEAVY CHAIN:

AB	**HCDR1**	**SEQ ID**	**HCDR2**	**SEQ ID**
100H8	DYNMD	NO:3	HINPNNGGILYNQKFKG	NO:10
74H10	DYNMD	NO:3	HINPNNGG**T**LYNQKFK**D**	NO:11
222E7	DYNMD	NO:3	HINPN**I**GG**T**LYNQKFKG	NO:12
55B3	DYNMD	NO:3	HINPN**T**GG**TI**YNQKFKG	NO:13

AB	**HCDR3**	**SEQ ID**
100H8	WAPLVRQPYWY	NO:20
74H10	WAPL**L**RQPYWY**FDV**	NO:21

222E7	WA**Q**L**Q**RQPYWY**FDV**	NO:22
55B3	WA**Q**L**L**RQPYWY**FDV**	NO:23

LIGHT CHAIN:

AB	LCDR1	SEQ ID	LCDR2	SEQ ID
100H8	KASQDINSYLN	NO:30	RADRLVD	NO:40
74H10	KASQDINSYLN	NO:30	RADRLVD	NO:40
222E7	KASQDINSYLN	NO:30	R**VN**RLVD	NO:38
55B3	KASQ**SVTNDVT**	NO:31	RA**N**RLVD	NO:39

AB	LCDR3	SEQ ID
100H8	LQYDEFPLT	NO:45
74H10	LQYDEFPLT	NO:45
222E7	LQYDEFPLT	NO:45
55B3	LQYDEFPLT	NO:45

Indeed, all of the HCDR1's in this group of four antibodies have the identical amino acid sequence (SEQ ID NO: 3). Whereas the four HCDR2's differ, they do not differ much. Notably, the HCDR2 of 100H8 differs from the three other HCDR2's by having an isoleucine residue at the ninth position rather than a threonine residue. 74H10 further differs from 100H8 by possessing an aspartic acid residue at its C-terminus rather than a glycine residue. Like 100H8, the other three HCDR2's have a glycine residue at their C-terminus. Unlike 100H8, the other three HCDR's have three additional amino acid residues (FDV) at their C-terminus. Both 74H10 and 55B3 have a leucine at their fourth position, rather than a valine residue like 100H8, whereas 222E7 has a glutamine residue. Finally, whereas both 100H8 and 74H10 have a proline residue at the third position, both 222E7 and 55B3 have a glutamine residue at the third position.

Three of the four LCDR1's in this group of four antibodies have the identical amino acid sequence (SEQ ID NO: 30), but whereas the LCDR1 of 55B3 shares their first four amino acid residues, its LCDR1 differs from the other three LCDR1s in its remaining seven amino acid residues. The LCDR2 of 100H8 and 74H10 have identical amino acid sequences (SEQ ID NO: 40), but the LCDR2 of 55B3 and 222E7 both differ from the other two LCDR2s by having an asparagine residue at position three instead of the aspartic acid residue of 100H8 and 74H10. In addition, the LCDR2 of 222E7 has a valine residue at position two rather than an alanine

residue found in the LCDR2 of the three other antibodies. Notably, all four LCDR3's in this group of four antibodies have the identical amino acid sequence (SEQ ID NO: 45).

The remaining six (6) antibodies to canine IL-31RA detailed above, *i.e.*, 65G9, 85C10, 224G3, 51F8, 209G5, and 218D9, can be further broken down into two pairs of antibodies that have noticeable identity in their respective CDR amino acid sequences, and two antibodies that are relative outliers. Accordingly, there is appreciable amino acid sequence identity between the sets of 6CDRs of antibody 65G9 with that of antibody 85C10 (*see*, Table 3). Consistently, antibodies 65G9 and 85C10 both bind two linear sequences in the C-terminal region of IL-31RA-ECD, one of which is the same (SEQ ID NO: 106; *see,* Table 6 and Figure 6D) and the other one has substantial overlap (compare SEQ ID NO: 107 with SEQ ID NO: 108; *see,* Table 6). Notably, one of the outliers, antibody 244G3, binds to an epitope comprising a single linear sequence in the C-terminal region of IL-31RA-ECD (*see*, Figure 6E), which is contained with the second linear sequence of antibody 85C10 (compare SEQ ID NO: 109 with SEQ ID NO: 108; *see,* Table 6).

The second group contains antibodies 51F8 and 209G5, which also have appreciable amino acid sequence identity between their respective sets of 6CDRs (*see*, Table 3). Consistently, antibodies 51F8 and 209G5 both bind two linear sequences in the N-terminal region of IL-31RA-ECD, one of which is the same (SEQ ID NO: 98; *see,* Table 6 and Figure 6B), whereas the second linear sequence of the IL-31RA-ECD that antibody 209G5 binds (SEQ ID NO: 101) is within the amino sequence of the second linear sequence of IL-31RA-ECD that antibody 51F8 binds (SEQ ID NO: 100; *see,* Table 6).

The other outlier, antibody 218D9 binds to two linear amino acid sequences located in the middle portion of the amino acid sequence of IL-31RA-ECD (SEQ ID NOs: 104 and 105; *see,* Figure 6C and Table 6). This antibody proved to be both a strong binder of IL-31RA and a good blocker of the binding of IL-31RA with IL-31.

EXAMPLE 5

BLOCKING ACTIVITY OF ANTI-IL-31 RECEPTOR *alpha* ANTIBODIES

The ability of anti-canine IL-31RA hybridoma supernatants to block the binding of IL-31 to IL-31RA were evaluated in the blocking ELISA described below.

Protocol

1) Coat 96-well half area plates with IL-31RA (1 µg/mL in PBS buffer), 25 µL/well.
2) Incubate the plates at 4°C overnight.

3) Wash the plates 3 times by PBST (PBS +0.05% Tween 20)

4) Block the plates with blocking buffer (PBS with 5% FBS), 25ul/well, for 30 minutes at room temperature.

5) Transfer 25 ul/well hybridoma supernatant to the 96-well plates, incubate 60 minutes at room temperature.

6) Wash the plates 3 times with PBST.

7) Transfer 25 µL/well of biotinylated IL31 (0.5 µg/mL in blocking buffer,) incubate 60 minutes at room temperature.

8) Wash the plates 3 times with PBST.

9) Add 25 µl/well Streptavidin-HRP, 1:5000 dilution in blocking buffer, to the plates and incubate 60 minutes at room temperature.

10) Wash the plates five times with PBST.

11) Add TMB based reagent to the plates for colorimetric reaction for 2-3 minutes.

12) Stop the reactions with 0.16 M sulfuric acid.

13) Read the plates by plate reader.

Results:

Out of the approximately 2000 hybridoma clones initially identified, approximately 300 were found to have binding affinity for IL-31RA, and about 10 of such clones also showed significant blocking of the binding of canine IL-31 to canine IL-31RA (*see,* Examples below).

EXAMPLE 6

FACS ASSAY FOR TESTING BLOCKING ACTIVITY OF MONOCLONAL ANTIBODIES AGAINST CANINE IL31-RA

In order to develop a cell-based assay to assess binding and blocking of canine IL-31 by anti-canine IL-31RA antibodies, the nucleotide sequences of the canine IL-31RA with c-terminal Flag tag and OSMR with c-terminal HA tag were prepared by chemical synthesis and then cloned into lentivirus vector Lenti-puro and Lenti-Hygro, respectively. The lentivirus Lenti-puro-IL31RA-Flag and Lenti-Hygro-OSMR-HA prepared from the Lenti-X 293T cells were co-transfected into CHO-k1 cells. The CHO stable cell pool co-expressing canine IL-31RA and OSMR was selected by FACS with anti-flag and anti-HA antibodies. Single cell clones were isolated from the stable pool. The developed CHO-IL-31RA/OSMR table cell line is applied to screen anti-canine IL-31RA monoclonal antibodies for blocking of IL-31 with its receptor complex IL-31RA/OSMR.

Materials

Cell line: CHO-IL-31RA/OSMR stable cell line

Cell growth medium: F-12K Medium with 10% FBS, 8μg/ml Puromycin and 200μg/ml hygromycin

Recombinant canine IL-31-His protein (0.5 μg/ml)

FACS Buffer: PBS

Isotype control: Mouse IgG (Genscript,A01007) (3 μg/ml)

Secondary antibody : Mouse anti-His tag antibody, 1μg/ml (Genscript, A01802)

Flow cytometer: BD FACSCanto

Flow Cytometry Procedure

1) CHO-IL-31RA/OSMR cells were grown in the growth medium in T75 flask.

2) Trypsinize to detach the cells, and then resuspend the cells in 5 mL fresh growth medium.

3) Spin down the cells at 300g for 3min, discard the supernatant and wash the cells twice with PBS.

4) Resuspend the cells in PBS as 2x106 cells/mL.

5) The cells were plated into 96-well assay plate with 50μl/well.

6) Mix anti-IL-31RA antibody (20μg/mL) or isotype control with IL-31 (1μg/ml), and then transfer 50μl of the mixture into each well of the assay plate.

7) After incubation at 4°C for 40min, the cells were washed twice by 150μl of cold PBS.

8) Add 100μl of the secondary antibody (1μg/mL) into each well of the assay plate.

9) After incubation at 4°C for 40min, the cells were washed twice by 150μl of cold PBS.

10) Resuspend the cells in 100μl/well cold PBS and read by the Flow cytometry.

As shown in Figure 3, the FACS result indicate that nine of the ten mouse anti-canine IL-31RA mAbs can significantly block the binding of IL-31 with the IL-31RA/OSMR complex presented on the CHO-IL-31RA/OSMR cells. The lead antibodies are 51F8, 74H10, 100H8, 209G5 and 218D9.

EXAMPLE 7

STAT-3 ASSAY

Stat-3 is known to be activated by IL-31 in cells comprising the the heterodimeric receptor for IL-31. In order to develop an assay to assess the activation of STAT-3 by canine IL-31, the nucleotide sequences encoding IL-31RA and OSMR, respectively, were prepared by

chemical synthesis and then cloned into expression vectors pcDNA3.1. The vectors containing the IL-31RA and OSMR nucleotide sequences, respectively, were co-transfected into Ba/f3 cells and the transfected cells, denoted as "Ba/f3-OI", were grown as a pool under antibiotic selection. The ability of canine IL-31 to induce STAT-3 activation was tested as follows.

Materials:

Cell line: Ba/f3-OI stable pool cells

Growth medium with mouse IL-3 or with canine IL-31 (cIL-31):

RPMI 1640 435 ml (ThermoFisher, 12633-020)

FBS 50 mL (SAFC cat# 12003c-500mL)

2-Mercaptoethanol (50 mM) 0.5 mL (Gibco 31350-010)

100X Pen Strep 5 mL (Gibco 15140-122 Lot1734040)

200mM L-Glu 10 ml (Gibco 25030-081 Lot1677185)

500 ng/mL Geneticin G418 (from Gibco or Sigma)

5 ng/mL mIL-3 or 100 ng/mL cIL-31

Starvation medium: the growth medium without mIL-3 and cIL-31

p-STAT3 (Tyr705) Assay Kit: PerkinElmer, ALSU-PST3-A-HV

Procedure:

Cell culture

1) Thaw a vial of the Ba/f3-OI cells, and grow the cells in the growth medium with mIL-3 in 37°C CO2 shaker with 125 rpm.

2) Passage the cells 2 – 3 passages to have the cells with $\geq$ 90% viability before set a cell-based assay.

3) To setup assay, harvest and resuspend the cells in the starvation medium to 1 x 10^7 viable cells/mL.

4) Dispense cells into 96-well plate, 50 µL/well (about 5 x 10^5 cells/well).

5) Three-fold dilute cIL-31 in starvation medium in a dilution plate, and then transfer 50 µL of each of the serial diluted cIL-31 aliquots into the cell plate.

6) Incubate the cell plate for 15-30 min in 37°C CO_2 shaker with 125 rpm for 1-2 hrs.

AlphaLISA assay as per manufacturer's instruction:

7) Spin down the cells, aspirate the supernatant, and add 1x lysis buffer of 50 – 100 µL/well. Incubate at RT for 10 min with 1000rpm shaking.

8) Remove 30 µL of the cell lysate into a ½ area plate or freeze and store at -80°C for future test.

SUREFIRE Assay:

8) Add 15 µL /well acceptor mix to the cell lysate. Seal and agitate plate for 2 min at 1000 rpm and then incubate for 1-2 hours at RT.

9) Add 15 µL/well donor mix to the cell lysate. Seal and agitate for 2 min at 1000 rpm, and then incubate for 1-2 hours at RT (the plate can be stored at 4°C overnight. Incubate at room temp for 1 hr before reading the plate next day)

10) Read the plate on Alpha plate reader at 520 – 620 nm.

Results:

Figure 4 shows the induction of STAT-3 phosphorylation by canine IL-31, which stimulates activation of STAT-3 in Ba/f3-OI cells in a dose dependent manner. Ba/f3 cells were used as the control. Ba/f3-OI cells expressing the IL-31 receptor complex were tested for IL-31-induced STAT-3 phosphorylation. The results indicate that STAT-3 phosphorylation was induced by IL-31 in the Baf3-OI cells in a dose-dependent manner, implying that: (i) the canine IL-31 receptor complex is successfully expressed on the cell surface; (ii) that the binding of canine IL-31 to the IL-31 receptor can stimulate the endogenous STAT3 phosphorylation; and (iii) then initiate its downstream signaling pathway.

EXAMPLE 8

BIOLOGICAL ACTIVITY OF ANTI-CANINE IL-31RA ANTIBODIES

The ability of the anti-canine IL-31RA mAbs to inhibit the activation of STAT-3 in Ba/f3-OI cells is assessed as follows:

1) Thaw a vial of the Ba/f3-OI cells, and grow the Ba/f3-OI cells in the growth medium with mIL-3 in 37°C CO2 shaker with 125 rpm.

2) Passage the cells 2 – 3 passages to have the cells with $\geq$ 90% viability before set a cell-based assay.

3) To setup assay, harvest and resuspend the cells in the starvation medium to 1 x 10^7 viable cells/mL.

4) Dispense cells into 96-well plate, 50 µL/well (about 5 x 10^5 cells/well).

5) Three-fold dilute the antibody in starvation medium in a row on a 96-well plate, starting concentration at 200 µg/mL. Then add 5µL cIL-31 in each well to get final concentration of 100 ng/mL.

6) Transfer 50 µL of the diluted antibody and cIL-31 mix into each well of the cell plate, gently mix.

7) Incubate the cell plate in 37°C CO_2 shaker with 125 rpm for 15-30 min. AlphaLISA assay as per manufacturer's instruction: (refer to Example 7)

As exemplified in Figure 5A for monoclonal antibodies 209G5, 218D9, and 85C10, in Figure 5B for monoclonal antibodies 100H8, 74H10, 51F8, and 85C10, and again in Figure 8 for various constructs of 218D9, all of the IL-31RA mAbs tested inhibit the canine IL-31 mediated STAT-3 phosphorylation in the Ba/f3-OI cells, whereas in the absence of these antibodies, there is no inhibition (labled IL-31). From Figure 8, the IC50 for the various 218D9 antibody constructs was calculated to be approximately: 2.2 nM for the chimeric antibody; 230 nM for c218D9VH3VL2; 8.3 nM for c218D9VH4VL2; 2.9 nM for c218D9VH4VL3; and 2.5 nM for c218D9VH3VL3.

EXAMPLE 9

IN VITRO BINDING OF ANTI-CANINE IL-31RA MONOCLONAL ANTIBODIES TO CANINE IL-31RA RECEPTOR

All kinetics measurements were performed by Octet HTX using SA biosensors and Data Acquisition 12.0 software. A biotin-labeled antigen (canine IL-31RA) was loaded onto the pre-rehydrated SA biosensors for 120s at a concentration of 1 μg/mL. Next, the biosensors were placed into Octet Kinetics Buffer (PBS+ 0.02% Tween 20, 0.1% BSA) for the blocking phase for 120s. Then, for the association phase, antigen loaded biosensors were placed into 2-fold serial dilutions from 100 nM down to 3.13 nM of anti-IL-31RA monoclonal antibody in Octet Kinetics Buffer for 300s. The last well was buffer alone and that sensor was used for reference sensor subtraction. Finally, the biosensors were placed into Octet Kinetics Buffer for the dissociation phase for 300s. Analysis was performed using Data Analysis 12.0 software and curves were fitted using a 1:1 binding model.

Binding affinity measurement results indicate that all the tested monoclonal antibodies have low nanomolar to sub-picomolar binding affinities ranging from about 1.5 nM to about 1 pM (*see*, Table 2 above). Two of the top blocking antibodies: 51F8 and 218D9, had KDs of about 0.2 nM and about 0.07 nM respectively (*see*, Table 5B below).

EXAMPLE 10

CANINIZED ANTIBODIES

The DNA and protein sequence of the canine heavy and light chains are known and can be obtained by searching of the NCBI gene and protein databases. As indicated above, for canine antibodies there are four known IgG subtypes: IgG-A, IgG-B, IgG-C, and IgG-D, and two types of light chains, *i.e.*, *kappa* and *lambda*. Without being bound by any specific approach, the process of producing caninized heavy and light chains that can be mixed in different combinations to produce caninized anti-canine IL-31 receptor *alpha* mAbs involves the following scheme:

Identify the DNA sequence of VH and VL domains comprising the CDRs of desired anti-IL-31 receptor *alpha* mAbs

i) Identify the H and L chain CDRs of desired anti-IL-31RA mAbs
ii) Identify a suitable sequence for H and L chain of canine IgG
iii) Identify the DNA sequence encoding the endogenous CDRs of canine IgG H and L chains of the above sequence.
iv) Replace the DNA sequence encoding endogenous canine H and L chain CDRs with DNA sequences encoding the desired anti-IL-31RA CDRs. In addition, optionally replace some canine framework residues with selected residues from the desired anti-IL-31 receptor alpha mAb framework regions.
v) Synthesize the DNA from step (v), clone it into a suitable expression plasmid, and transfect the plasmids containing desired caninized H and L chains into HEK 293 cells.
vi) Purify expressed caninized antibody from HEK 293 supernatant.
vii) Test purified caninized antibody for binding to canine IL-31 receptor *alpha* chain.

The application of the above outlined steps can result in a set of caninized H and L chain amino acid sequences provided below.

c51F8VH1-cIgGBm [SEQ ID NO: 72]

EVQLVQSGAEVKKPGASVKVSCKTSGYTFINTFIHWVRQAPGAGLDWMGQIDPANGNTKYAPKFQ
GRVTLTADTSTSTAYMELSSLRAGDIAVYYCARYYYVSSYFDVWGQGTLVTVSSASTTAPSVFPL
APSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVPSSRW
PSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTLLIAR
TPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKGKQFT
CKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEWQSNG
QQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHSPGK

c51F8VH2-cIgGBm [SEQ ID NO: 73]

EVQLVQSGAEVKKPGASVKVSCTASGFNIKNTFIHWVRQAPGAGLDWIGRIDPANGNTKYAPKFQ
GRVTLTADTSTSTAYMELSSLRAGDIAVYYCARYYYVSSYFDVWGQGTLVTVSSASTTAPSVFPL
APSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVPSSRW
PSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTLLIAR
TPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKGKQFT
CKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEWQSNG
QQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHSPGK

c51F8VH3-cIgGBm [SEQ ID NO: 74]

EVQLVQSGAEVKKPGASVKVSCTASGFNIKNTFIHWVRQAPGAGLDWIGRIDPANGNTKYAPKFQ
GRATLTADTSTNTAYMQLSSLRAGDIAVYYCARYYYVSSYFDVWGQGTLVTVSSASTTAPSVFPL
APSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVPSSRW
PSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTLLIAR
TPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKGKQFT
CKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEWQSNG
QQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHSPGK

c51F8VH4-cIgGBm [SEQ ID NO: 75]

EVQLVQSGAEVVKPGASVKVSCTASGFNIKNTFIHWVRQRPGAGLDWIGRIDPANGNTKYAPKFQ
GRATLTADTSTNTAYMQLSSLRAGDIAVYYCARYYYVSSYFDVWGTGTLVTVSSASTTAPSVFPL
APSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVPSSRW
PSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTLLIAR
TPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKGKQFT
CKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEWQSNG
QQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHSPGKR
NDAQPAVYLFQPSPDQLHTGSASVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYS
LSSTLTMSSTEYLSHELYSCEITHKSLPSTLIKSFQRSECQRVD

c51F8VL1-cCK [SEQ ID NO: 76]

EIVMTQSPASLSLSQEEKVTITCKASQSVTNDVTWYQQKPGQAPKLLIYSASNRYTGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCQQDYSSPWTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA

SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c51F8VL6-cCK [SEQ ID NO: 77]

DIVMTQTPLSLSVSPGETASISCKASQSVTNDVTWFRQKPGQSPQLLIYSASNRYTGVPDRFSGS
GSGTDFTLRISTVEADDTGVYYCQQDYSSPWTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c51F8VL7-cCK [SEQ ID NO: 78]

DIVMTQTPLSLSVSPGETASISCKASQSVTNDVTWFRQKPGQSPKLLIYSASNRYTGVPDRFSGS
GSGTDFTLRISTVEADDTGVYYCQQDYSSPWTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c100H8VH4-cIgGBm [SEQ ID NO: 79]

EVQLVQSGAEVKKPGASVKVSCKTSGYTFTDYNMDWVRQAPGAGLDWMGHINPNNGGILYNQKFK
GRVTLTADTSTSTAYMELSSLRAGDIAVYYCARWAPLVRQPYWYFDVWGQGTLVTVSSASTTAPS
VFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVP
SSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTL
LIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKG
KQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEW
QSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHS
PGK

c100H8VH7-cIgGBm [SEQ ID NO: 80]

EVQLVQSGAEVKKPGASVKVSCKASGYTFTDYNMDWVRQAPGAGLDWIGHINPNNGGILYNQKFK
GKATLTADTSTSTAYMELSSLRAGDIAVYYCARWAPLVRQPYWYFDVWGQGTLVTVSSASTTAPS
VFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVP
SSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTL
LIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKG
KQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEW

QSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHS
PGK

c100H8VH8-cIgGBm [SEQ ID NO: 81]

EVQLVQSGAEVKKPGASVKVSCKASGYTFTDYNMDWVRQAPGAGLDWIGHINPNNGGILYNQKFK
GKATLTVDRSTNTAYMELSSLRAGDIAVYFCARWAPLVRQPYWYFDVWGQGTLVTVSSASTTAPS
VFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTVP
SSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDTL
LIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLKG
KQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVEW
QSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSHS
PGK

c100H8VL1-cCK [SEQ ID NO: 82]

EIVMTQSPASLSLSQEEKVTITCKASQDINSYLNWYQQKPGQAPKLLIYRADRLVDGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCLQYDEFPLTFGAGTKVELKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c100H8VL2-cCK [SEQ ID NO: 83]

EIVMTQSPASLSLSQEEKVTITCKASQDINSYLNWFQQKPGQAPKLLIYRADRLVDGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCLQYDEFPLTFGAGTKLELKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c100H8VL3-cCK [SEQ ID NO: 84]

DIKMTQSPASLSLSQEEKVTITCKASQDINSYLNWFQQKPGQAPKLLIYRADRLVDGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCLQYDEFPLTFGAGTKLELKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c100H8VL4-cCK [SEQ ID NO: 85]

DIVMTQTPLSLSVSPGETASISCKASQDINSYLNWFRQKPGQSPQLLIYRADRLVDGVPDRFSGS
GSGTDFTLRISTVEADDTGVYYCLQYDEFPLTFGAGTKVELKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c218D9VH1-cIgGBm [SEQ ID NO: 86]

ELTLQESGPGLVKPSQTLSLTCVVSGGSVTTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRISITADTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSSASTTAP
SVFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTV
PSSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDT
LLIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLK
GKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVE
WQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSH
SPGK

c218D9VH2-cIgGBm [SEQ ID NO: 87]

ELTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRISITADTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSSASTTAP
SVFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTV
PSSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDT
LLIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLK
GKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVE
WQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSH
SPGK

c218D9VH3-cIgGBm [SEQ ID NO: 88]

EVTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRLSITKDTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSSASTTAP
SVFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTV
PSSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDT
LLIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLK
GKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVE

WQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSH
SPGK

c218D9VH4-cIgGBm [SEQ ID NO: 89]

EVTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRLSITKDTAKNQVFLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSSASTTAP
SVFPLAPSCGSTSGSTVALACLVSGYFPEPVTVSWNSGSLTSGVHTFPSVLQSSGLYSLSSMVTV
PSSRWPSETFTCNVAHPASKTKVDKPVPKRENGRVPRPPDCPKCPAPEMLGGPSVFIFPPKPKDT
LLIARTPEVTCVVVALDPEDPEVQISWFVDGKQMQTAKTQPREEQFAGTYRVVSVLPIGHQDWLK
GKQFTCKVNNKALPSPIERTISKARGQAHQPSVYVLPPSREELSKNTVSLTCLIKDFFPPDIDVE
WQSNGQQEPESKYRTTPPQLDEDGSYFLYSKLSVDKSRWQRGDTFICAVMHEALHNHYTQESLSH
SPGK

c218D9VL1-cCK [SEQ ID NO: 90]

EIVMTQSPASLSLSQEEKVTITCRASENIYSSLAWYQQKPGQAPKLLIYAATNLADGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCQHFRDTPPTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c218D9VL2-cCK [SEQ ID NO: 91]

DIVMTQTPLSLSVSPGETASISCRASENIYSSLAWFRQKPGQSPQLLIYAATNLADGVPDRFSGS
GSGTDFTLRISRVEADDTGVYYCQHFRDTPPTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c218D9VL3-cCK [SEQ ID NO: 92]

DIVMTQTPLSLSVSPGETASISCRASENIYSSLAWFRQKPGQSPQLLVYAATNLADGVPDRFSGS
GSGTDYTLRISRVEADDTGVYYCQHFRDTPPTFGQGTKLEIKRNDAQPAVYLFQPSPDQLHTGSA
SVVCLLNSFYPKDINVKWKVDGVIQDTGIQESVTEQDKDSTYSLSSTLTMSSTEYLSHELYSCEI
THKSLPSTLIKSFQRSECQRVD

c218D9VH1 [SEQ ID NO: 93]

ELTLQESGPGLVKPSQTLSLTCVVSGGSVTTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRISITADTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSS

c218D9VH2 [SEQ ID NO: 94]
ELTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRISITADTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSS

c218D9VH3 [SEQ ID NO: 95]
EVTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRLSITKDTAKNQFSLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSS

c218D9VH4 [SEQ ID NO: 96]
EVTLQESGPGLVKPSQTLSLTCSFSGFSLSTFGRGVGWIRQRPGRGLEWMGHIWWDDDKYYNPAL
KSRLSITKDTAKNQVFLQLSSMTTEDTAVYYCARIAGGLRRAPYAMDSWGQGTLVTVSS

c218D9VL1 [SEQ ID NO: 128]
EIVMTQSPASLSLSQEEKVTITCRASENIYSSLAWYQQKPGQAPKLLIYAATNLADGVPSRFSGS
GSGTDFSFTISSLEPEDVAVYYCQHFRDTPPTFGQGTKLEIK

c218D9VL2 [SEQ ID NO: 129]
DIVMTQTPLSLSVSPGETASISCRASENIYSSLAWFRQKPGQSPQLLIYAATNLADGVPDRFSGS
GSGTDFTLRISRVEADDTGVYYCQHFRDTPPTFGQGTKLEIK

c218D9VL3 [SEQ ID NO: 130]
DIVMTQTPLSLSVSPGETASISCRASENIYSSLAWFRQKPGQSPQLLVYAATNLADGVPDRFSGS
GSGTDYTLRISRVEADDTGVYYCQHFRDTPPTFGQGTKLEIK

EXAMPLE 11

REACTIVITY OF CANINIZED ANTIBODIES AGAINST CANINE IL-31RA

The caninized antibodies were tested for reactivity with canine IL-31RA as follows:

1. Coat 200 ng/well ofIL-31RA on an immunoplate and incubate the plate at 4°C overnight.
2. Wash the plate 3 times with phosphate buffered saline (PBS) containing 0.05% Tween 20 (PBST).

3. Block the plate with 0.5% bovine serum albumin (BSA) in PBS for 45 - 60 min at room temperature.

4. Wash the plate 3 times with PBST.

5. Three - fold dilute the caninized antibody in each column or row of dilution plate starting at 0. 3μg/mL.

6. Transfer the diluted caninized antibody into each column or row of the immunoplate, and incubate the plate for 45 - 60 min at room temperature.

7. Wash the plate 3 times with PBST.

8. Add 1 :4000 diluted horseradish peroxidase labeled anti - canine IgG Fc into each well of the plate, and then incubate the plate for 45 - 60 min at room temperature.

9. Wash the plate 3 times with PBST.

10. Add 3,3 ',5,5'-tetramethylbenzidine (TMB) Substrate into each well of the plate, and incubate the plate for 10 to 15 min at room temperature to develop the color.

11. Add 100 μL 1.5 M phosphoric acid into each well to stop the reaction. Read plate at 450nm with 540 nm reference wavelength.

As depicted in Figures 7A-7E, the caninized antibodies were tested for their reactivity to canine IL-31RA. The results indicate that the caninized antibodies have similar binding affinity as their corresponding parental antibodies (represented by their chimeric antibodies).

Table 5A below provides the relative binding affinities of the different caninized antibodies (EC50), relative to their corresponding mouse-canine chimera antibody (*see*, Figures 7A-7E). Although these are just relative numbers, caninized antibody 218D9 stands out as an antibody that has essentially the same binding affinity as its parental chimeric murine antibody.

The term "Chimeric" before the antibody number signifies that the antibody is a murine-canine chimeric antibody, *e.g.*, Chimeric 218D9 or Chimeric 51F8. In addition, an "m" before the antibody number followed by a "Chim" signifies that the antibody is a murine-canine chimeric antibody, *e.g.*, m224G3 Chim. The lower case "c" before the antibody number signifies that it is a caninized antibody, *e.g.*, c218D9VH4VL2.

TABLE 5A

RELATIVE BINDING AFFINITIES OF THE CANINIZED ANTIBODIES (EC50)

ANTIBODY	EC50
Chimeric 100H8	2.021
c100H8VH5L4	2.561
c100H8VH7L4	0.581

Chimeric 85C10	2.583
c85C10VH3L2	1.910
c85C10VH1L2	6.002
Chimeric 224G3	0.383
c224G3VH2L2	1.078
c224G3VH2L3	0.699
Chimeric 51F8	3.417
c51F8VH3L6	16.94
c51F8VH3L7	25.02
c51F8VH4L6	8.306
c51F8VH4L7	11.33
Chimeric 218D9	3.153
c218D9VH3L2	4.289
c218D9VH3L3	3.216
c218D9VH4L2	4.096
c218D9VH4L3	3.580

Table 5B below shows the binding constants of the caninized antibodies of 51F8 and 218D9. The results again indicate that caninized 218D9 antibodies have essentially the same binding affinity as their parental chimeric murine antibody, whereas caninized 51F8 antibodies have slightly weaker binding affinity than their parental chimeric murine antibody.

TABLE 5B

BINDING CONSTANTS OF THE CANINIZED 51F8 AND 218D9 ANTIBODIES

ANTIBODY	KD (M)	ka (1/Ms)	kdis (1/s)
chimeric 51F8	2.07E-10	6.84E+05	1.41E-04
c51F8VH3VL6	2.67E-09	4.28E+05	1.14E-03
c51F8VH3VL7	3.99E-09	4.01E+05	1.60E-03
c51F8VH4VL6	1.01E-09	4.45E+05	4.48E-04
c51F8VH4VL7	1.23E-09	4.16E+05	5.12E-04
chimeric 218D9	6.98E-11	6.64E+05	4.63E-05
c218D9VH3VL2	5.28E-10	6.41E+05	3.38E-04
c218D9VH3VL3	4.16E-11	6.61E+05	2.75E-05
c218D9VH4VL2	3.05E-10	5.41E+05	1.65E-04
c218D9VH4VL3	4.47E-11	6.01E+05	2.69E-05

EXAMPLE 12

MAPPING OF CANINE IL-31 RECEPTOR *alpha* EPITOPES USING MASS SPECTROSCOPY

A method based on chemical crosslinking and mass spectrometry detection was employed to identify epitopes recognized by anti-canine IL-31 receptor *alpha* mAbs [CovalX Instrument Incorporated, located at 999 Broadway, Suite 305, Saugus, MA 01906-4510]. The application of this technology to epitope mapping of canine IL-31 receptor *alpha* chain resulted in identification of epitopes recognized by the mAbs listed in Table 6 below. The results from the epitope mapping of canine IL-31 receptor *alpha* with the antibodies disclosed herein indicate that the mAbs recognize specific peptide epitopes that are present within the extracellular domain of canine IL-31 receptor *alpha* (*see,* Table 6 below). The results from the epitope mapping of canine IL-31RA with the eight antibodies included in Table 6, indicate that the mAbs recognize specific peptide epitopes that are present within the extracellular domain of canine IL-31RA**.** Notably**,** seven epitopes, which are distributed from N-terminus to C-terminus of IL-31RA-ECD were identified bound by the eight antibodies. Antibodies 100H8, 51F8, 209G5, and 55B3 share epitopes towards the N-terminus, while antibodies 65G9, 85C10 and 224G3 share epitopes towards the C-terminus of the IL-31RA-ECD. Antibody 218D9 has two unique epitopes located at middle portion of IL-31RA-ECD. As indicated by the functional results, all the eight monoclonal antibodies block IL-31 mediated STAT-3 phosphorylation, meaning that the seven epitopes are important in the interaction of canine IL-31RA with its ligand. This exhibits the complexity of the interaction of the canine IL-31 and canine IL-31 receptor complex. Five of the eight monoclonal antibodies tested were caninized, and the positions of their respective epitopes and their identified binding amino acid residues of the IL-31RA ECD are both presented in Figures 6A-6E and included in Table 6 below [*see also*, Figures 6A-6E which provides the epitopes on canine IL-31RA for the antibodies:100H8, 51F8, 218D9, 85C10, and 224G3, respectively, and the position of binding residues of the amino acid sequence of SEQ ID NO: 2 of the respective epitopes on the cIL-31R ECD antigen.]

TABLE 6
IL-31RA EPITOPES RECOGNIZED BY anti-IL-31RA MONOCLONAL

mAb	SEQ ID NO:	EPITOPE SEQUENCE	[1]RESIDUES IDENTIFIED AS BINDING
100H8	97	YWNLDAIMKIEPPEIFSVK	Y_{94}, K_{102}, and K_{112}

[1] Residue position on the cIL-31R ECD antigen with the amino acid sequence of SEQ ID NO: 2.

	103	RCAPAESMFWSGWSQEKVGT	R_{183},S_{193},and T_{202}
51F8	98 100	KIEPPEIFSVKSVLGIKR TEYVMTLRCAPAES	K_{102},S_{110},K_{112},K_{118},and R_{119} T_{176},Y_{178},and S_{189}
209G5	98 101	KIEPPEIFSVKSVLGIKR YVMTLR	
55B3	99 102	RPVLAPHSSTLKY RCAPAESM	
218D9	104 105	RRPVQLMWKKAT RIPALNEKT	R_{225} and T_{236} R_{298}, K_{305}, and T_{306}
65G9	106 107	RNGFIKNYTIFY RTSYSLQVMASTNAGGT	
85C10	106 108	RNGFIKNYTIFY SLQVMASTNAGGTNGT	R_{430},K_{435},T_{438}, and Y_{441} S_{472},T_{479}, and T_{487}
224G3	109	STNAGGTNGT	S_{478} and T_{487}

TABLE 7

SEQUENCES IN FIGURES 6A-6E

FIGURE	SEQ ID NO:	SEQUENCE
6A	119 120	SDITYWNLDAIMKIEPPEIFSVKSVLGIKRM MTLRCAPAESMFWSGWSQEKVGTTEEEAPYG
6B	121 122	IMKIEPPEIFSVKSVLGIKRM TELQAFTEYVMTLRCAPAESM
6C	123 124	AMVDGRRPVQLMWKKATGAPV GESPVATLRIPALNEKTFQCI
6D	125 126[#]	RNGFIKNYTIFYQAEDGKEFS SYSLQVMASTNAGGTNGTKIN
6E	126	SYSLQVMASTNAGGTNGTKIN

[#] Also in Figure 6E.

ST.25 SEQUENCE LISTING FROM PRIORITY FILING

SEQUENCE LISTING

<110> Intervet Inc.
Intervet International BV
Morsey, Mohamad
Zhang, Yuanzheng

<120> Caninized Antibodies To Canine Interleukin-31 Receptor Alpha II

<130> 25365-US-PSP

<160> 126

<170> PatentIn version 3.5

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<212> DNA
<213> Artificial Sequence

<220>
<223> canine with a His Tag

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tgcacctgga gccccgagaa ggaggccagc tacacctggt acaaggtgaa gagaacctac 120

agctacggct acaagagcga catctgcagc accgacaaca gcaccagagg caaccacgcc 180

agctgcagct tcctgccccc caccatcacc aaccccgaca actacaccat ccaggtggag 240

gcccagaacg ccgacggcat catgaagagc gacatcacct actggaacct ggacgccatc 300

atgaagatcg agcccccccga gatcttcagc gtgaagagcg tgctgggcat caagagaatg 360

ctgcagatca agtggatcag acccgtgctg gccccccaca gcagcaccct gaagtacacc 420

ctgagattca gaaccatcaa cagcgcctac tggatggagg tgaacttcac caaggaggac 480

atcgacagag acgagaccta caacctgacc gagctgcagg ccttcaccga gtacgtgatg 540

accctgagat gcgccccccgc cgagagcatg ttctggagcg gctggagcca ggagaaggtg 600

ggcaccaccg aggaggaggc cccctacggc ctggacctgt ggagagtgct gaagcccgcc 660

atggtggacg gcagaagacc cgtgcagctg atgtggaaga aggccaccgg cgcccccgtg 720

ctggagaagg ccctgggcta caacatctgg tacttccccg agaacaacac caacctgacc 780

gagaccgtga acaccaccaa ccagacccac gagctgtacc tgggcggcaa gacctactgg 840

gtgtacgtgg tgagctacaa cagcctgggc gagagccccg tggccaccct gagaatcccc 900

gccctgaacg agaagacctt ccagtgcatc gaggccatgc aggcctgcct gacccaggac 960

cagctggtgg tggagtggca gagcagcgcc cccgaggtgg acacctggat ggtggagtgg 1020

ttccccgacg tggacagcga gcccagcagc ttcagctggg agagcgtgag ccaggccaga 1080

aactggacca tccagaagga cgagctgaag cccctgtggt gctacaacat cagcgtgtac 1140

cccgtgctga gagacagagt gggccagccc tacagcaccc aggcctacgt gcaggagggc 1200

atccccagcg ccggccccgt gacccaggcc gacagcatcg gcgtgaagac cgtgaccatc 1260

acctggaagg agatccccaa gagcaagaga aacggcttca tcaagaacta caccatcttc 1320

taccaggccg aggacggcaa ggagttcagc aagaccgtga acagcaacat cctgcagtac 1380

agactggaga gcctgaccag aagaaccagc tacagcctgc aggtgatggc cagcaccaac 1440

gccggcggca ccaacggcac caagatcaac ttcaagaccc tgagcatcag ccaccaccac 1500

caccaccacc accaccacca c 1521

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<211> 507
<212> PRT
<213> Artificial Sequence

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<223> canine with a His Tag

<400> 2

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Glu Asn Phe Thr Cys Thr Trp Ser Pro Glu Lys Glu Ala Ser Tyr Thr
20 25 30

Trp Tyr Lys Val Lys Arg Thr Tyr Ser Tyr Gly Tyr Lys Ser Asp Ile
35 40 45

Cys Ser Thr Asp Asn Ser Thr Arg Gly Asn His Ala Ser Cys Ser Phe
50 55 60

Leu Pro Pro Thr Ile Thr Asn Pro Asp Asn Tyr Thr Ile Gln Val Glu
65 70 75 80

Ala Gln Asn Ala Asp Gly Ile Met Lys Ser Asp Ile Thr Tyr Trp Asn
85 90 95

Leu Asp Ala Ile Met Lys Ile Glu Pro Pro Glu Ile Phe Ser Val Lys
100 105 110

Ser Val Leu Gly Ile Lys Arg Met Leu Gln Ile Lys Trp Ile Arg Pro
115 120 125

Val Leu Ala Pro His Ser Ser Thr Leu Lys Tyr Thr Leu Arg Phe Arg
130 135 140

Thr Ile Asn Ser Ala Tyr Trp Met Glu Val Asn Phe Thr Lys Glu Asp
145 150 155 160

Ile Asp Arg Asp Glu Thr Tyr Asn Leu Thr Glu Leu Gln Ala Phe Thr
165 170 175

Glu Tyr Val Met Thr Leu Arg Cys Ala Pro Ala Glu Ser Met Phe Trp
180 185 190

Ser Gly Trp Ser Gln Glu Lys Val Gly Thr Thr Glu Glu Glu Ala Pro
195 200 205

Tyr Gly Leu Asp Leu Trp Arg Val Leu Lys Pro Ala Met Val Asp Gly
210 215 220

Arg Arg Pro Val Gln Leu Met Trp Lys Lys Ala Thr Gly Ala Pro Val
225 230 235 240

Leu Glu Lys Ala Leu Gly Tyr Asn Ile Trp Tyr Phe Pro Glu Asn Asn
245 250 255

Thr Asn Leu Thr Glu Thr Val Asn Thr Thr Asn Gln Thr His Glu Leu
260 265 270

Tyr Leu Gly Gly Lys Thr Tyr Trp Val Tyr Val Val Ser Tyr Asn Ser
275 280 285

Leu Gly Glu Ser Pro Val Ala Thr Leu Arg Ile Pro Ala Leu Asn Glu

290 295 300

Lys Thr Phe Gln Cys Ile Glu Ala Met Gln Ala Cys Leu Thr Gln Asp
305 310 315 320

Gln Leu Val Val Glu Trp Gln Ser Ser Ala Pro Glu Val Asp Thr Trp
325 330 335

Met Val Glu Trp Phe Pro Asp Val Asp Ser Glu Pro Ser Ser Phe Ser
340 345 350

Trp Glu Ser Val Ser Gln Ala Arg Asn Trp Thr Ile Gln Lys Asp Glu
355 360 365

Leu Lys Pro Leu Trp Cys Tyr Asn Ile Ser Val Tyr Pro Val Leu Arg
370 375 380

Asp Arg Val Gly Gln Pro Tyr Ser Thr Gln Ala Tyr Val Gln Glu Gly
385 390 395 400

Ile Pro Ser Ala Gly Pro Val Thr Gln Ala Asp Ser Ile Gly Val Lys
405 410 415

Thr Val Thr Ile Thr Trp Lys Glu Ile Pro Lys Ser Lys Arg Asn Gly
420 425 430

Phe Ile Lys Asn Tyr Thr Ile Phe Tyr Gln Ala Glu Asp Gly Lys Glu
435 440 445

Phe Ser Lys Thr Val Asn Ser Asn Ile Leu Gln Tyr Arg Leu Glu Ser
450 455 460

Leu Thr Arg Arg Thr Ser Tyr Ser Leu Gln Val Met Ala Ser Thr Asn
465 470 475 480

Ala Gly Gly Thr Asn Gly Thr Lys Ile Asn Phe Lys Thr Leu Ser Ile
485 490 495

Ser His His His His His His His His His His
500 505

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<213> Mus musculus

<400> 3

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1 5

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1 5

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<212> PRT
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<400> 5

Asn Thr Phe Ile His
1 5

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<400> 6

Asn Thr Tyr Met His
1 5

<210> 7
<211> 5
<212> PRT
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<400> 7

Asp Tyr Gly Met Asn
1 5

<210> 8
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<213> Mus musculus

<400> 8

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1 5

<210> 9
<211> 5
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<213> Mus musculus

<400> 9

Ser Tyr Gly Met Ser
1 5

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<211> 17
<212> PRT
<213> Mus musculus

<400> 10

His Ile Asn Pro Asn Asn Gly Gly Ile Leu Tyr Asn Gln Lys Phe Lys
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Gly

<210> 11
<211> 17
<212> PRT
<213> Mus musculus

<400> 11

His Ile Asn Pro Asn Asn Gly Gly Thr Leu Tyr Asn Gln Lys Phe Lys
1 5 10 15

Asp

<210> 12
<211> 17
<212> PRT
<213> Mus musculus

<400> 12

His Ile Asn Pro Asn Ile Gly Gly Thr Leu Tyr Asn Gln Lys Phe Lys
1 5 10 15

Gly

<210> 13
<211> 17
<212> PRT
<213> Mus musculus

<400> 13

His Ile Asn Pro Asn Thr Gly Gly Thr Ile Tyr Asn Gln Lys Phe Lys
1 5 10 15

Gly

<210> 14
<211> 17
<212> PRT
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<400> 14

Val Ile Asn Pro Asn Tyr Gly Thr Ser Ser His Asn Gln Lys Phe Lys
1 5 10 15

Gly

<210> 15
<211> 17
<212> PRT
<213> Mus musculus

<400> 15

Arg Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe Gln
1 5 10 15

Gly

<210> 16
<211> 17
<212> PRT
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<400> 16

Arg Ile Asp Pro Ala Asn Gly Asn Ser Lys Tyr Ala Pro Lys Phe Gln
1 5 10 15

Gly

<210> 17
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<212> PRT
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<400> 17

Ile Ile Asn Pro Asn Tyr Gly Thr Ala Ser Ser Asn Pro Lys Phe Lys
1 5 10 15

Asp

<210> 18
<211> 16
<212> PRT
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<400> 18

His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala Leu Lys Ser
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<210> 19
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<400> 19

Thr Ile Ser Asn Gly Gly Ser Tyr Thr Tyr Tyr Pro Asp Ser Val Lys
1 5 10 15

Gly

<210> 20
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<212> PRT
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1 5 10

<210> 21
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1 5 10

<210> 22
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<400> 22

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1 5 10

<210> 23
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<400> 23

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1 5 10

<210> 24
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<212> PRT
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<400> 24

Ala Leu Asp Asp Tyr Tyr Phe Tyr Trp Tyr
1 5 10

<210> 25
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<212> PRT
<213> Mus musculus

<400> 25

Tyr Tyr Tyr Val Ser Ser Tyr
1 5

<210> 26
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<213> Mus musculus

<400> 26

Tyr Tyr Tyr Val Ser Ser His Phe Asp Val
1 5 10

<210> 27
<211> 13
<212> PRT
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<400> 27

Ala Phe Asp Gly Tyr Tyr Phe Tyr Trp Tyr Phe Asp Val
1 5 10

<210> 28
<211> 14
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<400> 28

Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp Ser
1 5 10

<210> 29

<211> 6
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<213> Mus musculus

<400> 29

Asn Glu Pro Pro Asp Tyr
1 5

<210> 30
<211> 11
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<400> 30

Lys Ala Ser Gln Asp Ile Asn Ser Tyr Leu Asn
1 5 10

<210> 31
<211> 11
<212> PRT
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<400> 31

Lys Ala Ser Gln Asp Ile Asn Ser Tyr Phe Asn
1 5 10

<210> 32
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<400> 32

Lys Ala Ser Gln Ser Val Thr Asn Asp Val Thr
1 5 10

<210> 33
<211> 16
<212> PRT
<213> Mus musculus

<400> 33

Arg Ser Ser Gln Ser Leu Val His Thr Asn Gly Asn Thr Tyr Leu His
1 5 10 15

<210> 34
<211> 16
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<213> Mus musculus

<400> 34

Arg Ser Ser Gln Ser Leu Ile His Thr Asn Gly Asn Thr Tyr Leu His
1 5 10 15

<210> 35
<211> 11
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<213> Mus musculus

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Arg Ala Ser Glu Asn Ile Tyr Ser Ser Leu Ala
1 5 10

<210> 36
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<400> 36

Thr Ala Ser Ser Ser Val Gly Ser Ser Tyr Leu His
1 5 10

<210> 37
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<400> 37

Lys Val Ser Asn Arg Phe Ser
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<210> 38
<211> 7
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1 5

<210> 39
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<400> 39

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1 5

<210> 40
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<400> 40

Arg Ala Asp Arg Leu Val Asp
1 5

<210> 41
<211> 7
<212> PRT
<213> Mus musculus

<400> 41

Ser Ala Ser Asn Arg Tyr Thr
1 5

<210> 42
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<400> 42

Tyr Ala Ser Asn Arg Tyr Thr
1 5

<210> 43
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<400> 43

Ala Ala Thr Asn Leu Ala Asp
1 5

<210> 44
<211> 7
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<400> 44

Asp Thr Ser Asn Leu Ala Ser
1 5

<210> 45
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<212> PRT
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Leu Gln Tyr Asp Glu Phe Pro Leu Thr
1 5

<210> 46
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Ser Gln Thr Thr His Val Pro Leu Thr
1 5

<210> 47
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1 5

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<211> 9
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<400> 48

Gln Gln Asp Tyr Ser Ser Pro Phe Thr

1 5

<210> 49
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His Gln Tyr His Arg Ser Pro Tyr Thr
1 5

<210> 50
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<212> PRT
<213> Mus musculus

<400> 50

Ser Gln Ser Thr His Val Pro Leu Thr
1 5

<210> 51
<211> 9
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<213> Mus musculus

<400> 51

Gln His Phe Arg Asp Thr Pro Pro Thr
1 5

<210> 52
<211> 123
<212> PRT
<213> Mus musculus

<400> 52

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1 5 10 15

Ser Val Lys Ile Pro Cys Lys Ala Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Lys Gln Ser His Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Asn Gly Gly Ile Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Val Asp Arg Ser Ser Asn Thr Ala Tyr
65 70 75 80

Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Thr Ala Val Tyr Phe Cys
85 90 95

Ala Arg Trp Ala Pro Leu Val Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Thr Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 53
<211> 107
<212> PRT
<213> Mus musculus

<400> 53

Asp Ile Lys Met Thr Gln Ser Pro Ser Ser Ile Phe Ala Ser Leu Gly
1 5 10 15

Glu Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Gln Gln Lys Pro Gly Lys Ser Pro Lys Thr Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Gln Asp Tyr Ser Leu Thr Ile Ser Ser Leu Glu Tyr
65 70 75 80

Glu Asp Met Gly Ile Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Asn
100 105

<210> 54
<211> 123
<212> PRT
<213> Mus musculus

<400> 54

Glu Val Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ser
1 5 10 15

Ser Val Lys Ile Pro Cys Lys Ala Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Lys Gln Ser His Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Ile Gly Gly Thr Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Ser Val Asp Lys Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Trp Ala Gln Leu Gln Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Pro Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 55
<211> 107
<212> PRT
<213> Mus musculus

<400> 55

Asp Ile Lys Met Thr Gln Ser Pro Ser Ser Met Tyr Ala Ser Leu Gly
1 5 10 15

Glu Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Gln Gln Lys Pro Gly Lys Ser Pro Lys Thr Leu Ile
35 40 45

Tyr Arg Val Asn Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Gln Asp Tyr Ser Leu Thr Ile Ser Ser Leu Glu Tyr
65 70 75 80

Glu Asp Met Gly Ile Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys
100 105

<210> 56
<211> 123
<212> PRT
<213> Mus musculus

<400> 56

Glu Val Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Ile Pro Cys Lys Ala Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Lys Gln Ser His Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Thr Gly Gly Thr Ile Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Val Asp Lys Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Asp Leu Arg Ser Leu Thr Ser Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Trp Ala Gln Leu Leu Arg Gln Pro Tyr Trp Tyr Phe Asp Val

100 105 110

Trp Gly Thr Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 57
<211> 107
<212> PRT
<213> Mus musculus

<400> 57

Asp Ile Lys Met Thr Gln Ser Pro Ser Ser Met Tyr Ala Ser Leu Gly
1 5 10 15

Glu Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Phe Asn Trp Val Gln Gln Lys Pro Gly Lys Ser Pro Lys Thr Leu Ile
35 40 45

Phe Arg Ala Asn Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Gln Asp Tyr Ser Leu Thr Ile Asn Ser Leu Glu Tyr
65 70 75 80

Glu Asp Met Gly Ile Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys
100 105

<210> 58
<211> 123
<212> PRT
<213> Mus musculus

<400> 58

Glu Val Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Met Lys Ile Pro Cys Lys Thr Ser Gly Tyr Thr Phe Thr Asp Tyr

20 25 30

Asn Met Asp Trp Val Lys Gln Ser His Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Asn Gly Gly Thr Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Asp Lys Ala Thr Leu Thr Val Asp Arg Ser Ser Asn Thr Ala Tyr
65 70 75 80

Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Trp Ala Pro Leu Leu Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Thr Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 59
<211> 107
<212> PRT
<213> Mus musculus

<400> 59

Asp Ile Lys Met Thr Gln Ser Pro Ser Ser Met Tyr Ala Ser Leu Gly
1 5 10 15

Glu Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Gln Gln Lys Pro Gly Lys Ser Pro Lys Thr Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Gln Asp Tyr Ser Leu Thr Ile Ser Ser Leu Glu Tyr
65 70 75 80

Glu Asp Met Gly Ile Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Asp Gly Thr Lys Leu Glu Leu Asn
100 105

<210> 60
<211> 119
<212> PRT
<213> Mus musculus

<400> 60

Glu Val Gln Leu Gln Gln Ser Val Ala Glu Leu Val Arg Pro Gly Ala
1 5 10 15

Ser Val Lys Leu Ser Cys Thr Ala Ser Gly Phe Asn Ile Lys Asn Thr
20 25 30

Tyr Met His Trp Val Lys Glu Arg Pro Glu Gln Gly Leu Glu Trp Ile
35 40 45

Gly Arg Ile Asp Pro Ala Asn Gly Asn Ser Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Lys Ala Thr Ile Thr Thr Asp Thr Ser Ser Asn Thr Ala Tyr
65 70 75 80

Leu Gln Leu Ser Ser Leu Thr Ser Glu Asp Thr Ala Ile Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser His Phe Asp Val Trp Gly Thr Gly
100 105 110

Thr Thr Val Thr Val Ser Ser
115

<210> 61
<211> 107
<212> PRT
<213> Mus musculus

<400> 61

Ser Ile Val Met Thr Gln Thr Pro Lys Phe Leu Leu Val Ser Ala Gly
1 5 10 15

Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Thr Asn Asp
20 25 30

Val Thr Trp Tyr Gln Gln Lys Pro Gly Gln Ser Pro Lys Leu Leu Ile
35 40 45

Tyr Tyr Ala Ser Asn Arg Tyr Thr Gly Val Pro Asp Arg Phe Thr Gly
50 55 60

Ser Gly Tyr Gly Thr Asp Phe Thr Phe Thr Ile Ser Thr Val Gln Ala
65 70 75 80

Glu Asp Leu Ala Val Tyr Phe Cys Gln Gln Asp Tyr Ser Ser Pro Phe
85 90 95

Thr Phe Gly Ser Gly Thr Lys Leu Glu Ile Lys
100 105

<210> 62
<211> 119
<212> PRT
<213> Mus musculus

<400> 62

Glu Val Gln Leu Gln Gln Ser Val Ala Glu Leu Val Arg Pro Gly Ala
1 5 10 15

Ser Val Lys Leu Ser Cys Thr Ala Ser Gly Phe Asn Ile Lys Asn Thr
20 25 30

Phe Ile His Trp Val Lys Gln Arg Pro Glu Gln Gly Leu Glu Trp Ile
35 40 45

Gly Arg Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Arg Ala Thr Leu Thr Ala Asp Thr Ser Ser Asn Thr Ala Tyr
65 70 75 80

Leu Gln Leu Ser Ser Leu Thr Ser Glu Asp Thr Ala Ile Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser Tyr Phe Asp Val Trp Gly Thr Gly
100 105 110

Thr Thr Val Thr Val Ser Ser
115

<210> 63
<211> 107
<212> PRT
<213> Mus musculus

<400> 63

Ser Ile Val Met Thr Gln Thr Pro Lys Phe Leu Leu Val Ser Ala Gly
1 5 10 15

Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Thr Asn Asp
20 25 30

Val Thr Trp Tyr Gln Gln Lys Pro Gly Gln Ser Pro Lys Leu Leu Ile
35 40 45

Phe Ser Ala Ser Asn Arg Tyr Thr Gly Val Pro Asp Arg Phe Thr Gly
50 55 60

Ser Gly Tyr Gly Thr Asp Phe Thr Phe Thr Ile Ser Thr Val Gln Ala
65 70 75 80

Glu Asp Leu Ala Val Tyr Phe Cys Gln Gln Asp Tyr Ser Ser Pro Trp
85 90 95

Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys
100 105

<210> 64
<211> 122
<212> PRT
<213> Mus musculus

<400> 64

Glu Phe Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Ser Phe Thr Asp Tyr
20 25 30

Gly Met Asn Trp Leu Lys Gln Ile Asn Gly Lys Ser Leu Glu Trp Ile
35 40 45

Ala Ile Ile Asn Pro Asn Tyr Gly Thr Ala Ser Ser Asn Pro Lys Phe
50 55 60

Lys Asp Lys Ala Thr Leu Thr Val Asp His Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Gln Leu Ser Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Ala Phe Asp Gly Tyr Tyr Phe Tyr Trp Tyr Phe Asp Val Trp
100 105 110

Gly Thr Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 65
<211> 112
<212> PRT
<213> Mus musculus

<400> 65

Asp Val Val Met Thr Gln Thr Pro Leu Ser Leu Pro Val Ser Leu Gly
1 5 10 15

Asp Gln Ala Ser Ile Ser Cys Arg Ser Ser Gln Ser Leu Ile His Thr
20 25 30

Asn Gly Asn Thr Tyr Leu His Trp Tyr Leu Gln Lys Pro Gly Gln Ser
35 40 45

Pro Lys Leu Leu Ile Tyr Lys Val Ser Asn Arg Phe Ser Gly Val Pro
50 55 60

Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Lys Ile
65 70 75 80

Ser Arg Leu Glu Ala Glu Asp Leu Gly Val Tyr Phe Cys Ser Gln Ser
85 90 95

Thr His Val Pro Leu Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Arg
100 105 110

<210> 66
<211> 122
<212> PRT
<213> Mus musculus

<400> 66

Glu Phe Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Ile Ser Cys Lys Ala Ser Gly Tyr Ser Phe Thr Asp Tyr
20 25 30

Ser Ile Asn Trp Val Lys Gln Ser Asn Gly Lys Ser Leu Glu Trp Ile
35 40 45

Gly Val Ile Asn Pro Asn Tyr Gly Thr Ser Ser His Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Met Thr Val Asp Gln Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Gln Leu Asn Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Ala Leu Asp Asp Tyr Tyr Phe Tyr Trp Tyr Phe Asp Val Trp
100 105 110

Gly Ile Gly Thr Thr Val Thr Val Ser Ser
115 120

<210> 67

<211> 112
<212> PRT
<213> Mus musculus

<400> 67

Asp Val Val Met Thr Gln Thr Pro Leu Ser Leu Pro Val Ser Leu Gly
1 5 10 15

Asp Gln Ala Ser Ile Ser Cys Arg Ser Ser Gln Ser Leu Val His Thr
20 25 30

Asn Gly Asn Thr Tyr Leu His Trp Tyr Leu Gln Lys Pro Gly Gln Ser
35 40 45

Pro Lys Leu Leu Ile Tyr Lys Val Ser Asn Arg Phe Ser Gly Val Pro
50 55 60

Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Lys Ile
65 70 75 80

Ser Arg Val Glu Ala Glu Asp Leu Gly Val Tyr Phe Cys Ser Gln Thr
85 90 95

Thr His Val Pro Leu Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys
100 105 110

<210> 68
<211> 124
<212> PRT
<213> Mus musculus

<400> 68

Gln Val Thr Leu Lys Glu Ser Gly Pro Gly Ile Leu Gln Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Pro Ser Gly Lys Gly Leu Asp
35 40 45

Trp Leu Thr His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala

50 55 60

Leu Lys Ser Arg Leu Thr Ile Ser Lys Asp Thr Ser Lys Asn Gln Val
65 70 75 80

Phe Leu Lys Ile Ala Asn Val Asp Thr Ala Asp Thr Ala Thr Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Ser Val Thr Val Ser Ser
115 120

<210> 69
<211> 107
<212> PRT
<213> Mus musculus

<400> 69

Asp Ile Gln Met Thr Gln Ser Pro Ala Ser Leu Ser Val Ser Val Gly
1 5 10 15

Glu Thr Val Thr Ile Thr Cys Arg Ala Ser Glu Asn Ile Tyr Ser Ser
20 25 30

Leu Ala Trp Tyr Gln Gln Lys Gln Gly Lys Ser Pro Gln Leu Leu Val
35 40 45

Tyr Ala Ala Thr Asn Leu Ala Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Gln Tyr Ser Leu Lys Ile Asn Ser Leu Gln Ser
65 70 75 80

Glu Asp Ser Gly Asn Tyr Tyr Cys Gln His Phe Arg Asp Thr Pro Pro
85 90 95

Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys
100 105

<210> 70
<211> 115
<212> PRT
<213> Mus musculus

<400> 70

Glu Val Gln Leu Val Glu Ser Gly Gly Asp Leu Val Lys Pro Gly Gly
1 5 10 15

Ser Leu Lys Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Ser Tyr
20 25 30

Gly Met Ser Trp Val Arg Gln Thr Pro Asp Lys Arg Leu Glu Trp Val
35 40 45

Ala Thr Ile Ser Asn Gly Gly Ser Tyr Thr Tyr Tyr Pro Asp Ser Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ala Lys Asn Thr Leu Tyr
65 70 75 80

Met Gln Met Ser Ser Leu Lys Ser Glu Asp Thr Ala Met Tyr Tyr Cys
85 90 95

Ala Arg Asn Glu Pro Pro Asp Tyr Trp Gly Gln Gly Thr Ser Val Thr
100 105 110

Val Ser Ser
115

<210> 71
<211> 108
<212> PRT
<213> Mus musculus

<400> 71

Gln Ile Val Leu Thr Gln Ser Pro Ala Ile Met Ser Ala Ser Leu Gly
1 5 10 15

Glu Arg Val Thr Met Thr Cys Thr Ala Ser Ser Ser Val Gly Ser Ser
20 25 30

Tyr Leu His Trp Tyr Gln Gln Lys Pro Gly Ser Ser Pro Lys Leu Trp
35 40 45

Ile Tyr Asp Thr Ser Asn Leu Ala Ser Gly Val Pro Val Arg Phe Ser
50 55 60

Gly Ser Gly Ser Gly Thr Ser Tyr Ser Leu Thr Ile Ser Ser Met Glu
65 70 75 80

Ala Glu Asp Ala Ala Thr Tyr Tyr Cys His Gln Tyr His Arg Ser Pro
85 90 95

Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys
100 105

<210> 72
<211> 454
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 72

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Thr Ser Gly Tyr Thr Phe Ile Asn Thr
20 25 30

Phe Ile His Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Met
35 40 45

Gly Gln Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Arg Val Thr Leu Thr Ala Asp Thr Ser Thr Ser Thr Ala Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser Tyr Phe Asp Val Trp Gly Gln Gly
100 105 110

Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala Pro Ser Val Phe
115 120 125

Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser Thr Val Ala Leu
130 135 140

Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val Thr Val Ser Trp
145 150 155 160

Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe Pro Ser Val Leu
165 170 175

Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val Thr Val Pro Ser
180 185 190

Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val Ala His Pro Ala
195 200 205

Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg Glu Asn Gly Arg
210 215 220

Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala Pro Glu Met Leu
225 230 235 240

Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Thr Leu
245 250 255

Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Ala Leu Asp
260 265 270

Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys Gln
275 280 285

Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Ala Gly Thr
290 295 300

Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu Lys
305 310 315 320

Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser Pro
325 330 335

Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro Ser
340 345 350

Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr Val
355 360 365

Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp Val
370 375 380

Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg Thr
385 390 395 400

Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser Lys
405 410 415

Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile Cys
420 425 430

Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser Leu
435 440 445

Ser His Ser Pro Gly Lys
450

<210> 73
<211> 454
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 73

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Thr Ala Ser Gly Phe Asn Ile Lys Asn Thr
20 25 30

Phe Ile His Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Ile
35 40 45

Gly Arg Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Arg Val Thr Leu Thr Ala Asp Thr Ser Thr Ser Thr Ala Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser Tyr Phe Asp Val Trp Gly Gln Gly
100 105 110

Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala Pro Ser Val Phe
115 120 125

Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser Thr Val Ala Leu
130 135 140

Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val Thr Val Ser Trp
145 150 155 160

Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe Pro Ser Val Leu
165 170 175

Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val Thr Val Pro Ser
180 185 190

Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val Ala His Pro Ala
195 200 205

Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg Glu Asn Gly Arg
210 215 220

Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala Pro Glu Met Leu
225 230 235 240

Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Thr Leu
245 250 255

Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Ala Leu Asp
260 265 270

Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys Gln
275 280 285

Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Ala Gly Thr
290 295 300

Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu Lys
305 310 315 320

Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser Pro
325 330 335

Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro Ser
340 345 350

Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr Val
355 360 365

Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp Val
370 375 380

Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg Thr
385 390 395 400

Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser Lys
405 410 415

Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile Cys
420 425 430

Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser Leu
435 440 445

Ser His Ser Pro Gly Lys
450

<210> 74
<211> 454
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 74

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Thr Ala Ser Gly Phe Asn Ile Lys Asn Thr
20 25 30

Phe Ile His Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Ile
35 40 45

Gly Arg Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Arg Ala Thr Leu Thr Ala Asp Thr Ser Thr Asn Thr Ala Tyr
65 70 75 80

Met Gln Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser Tyr Phe Asp Val Trp Gly Gln Gly
100 105 110

Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala Pro Ser Val Phe
115 120 125

Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser Thr Val Ala Leu
130 135 140

Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val Thr Val Ser Trp
145 150 155 160

Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe Pro Ser Val Leu
165 170 175

Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val Thr Val Pro Ser
180 185 190

Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val Ala His Pro Ala
195 200 205

Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg Glu Asn Gly Arg
210 215 220

Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala Pro Glu Met Leu
225 230 235 240

Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Thr Leu
245 250 255

Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Ala Leu Asp
260 265 270

Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys Gln
275 280 285

Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Ala Gly Thr
290 295 300

Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu Lys
305 310 315 320

Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser Pro
325 330 335

Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro Ser
340 345 350

Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr Val
355 360 365

Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp Val
370 375 380

Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg Thr
385 390 395 400

Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser Lys
405 410 415

Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile Cys
420 425 430

Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser Leu
435 440 445

Ser His Ser Pro Gly Lys
450

<210> 75
<211> 564
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 75

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Thr Ala Ser Gly Phe Asn Ile Lys Asn Thr
20 25 30

Phe Ile His Trp Val Arg Gln Arg Pro Gly Ala Gly Leu Asp Trp Ile
35 40 45

Gly Arg Ile Asp Pro Ala Asn Gly Asn Thr Lys Tyr Ala Pro Lys Phe
50 55 60

Gln Gly Arg Ala Thr Leu Thr Ala Asp Thr Ser Thr Asn Thr Ala Tyr
65 70 75 80

Met Gln Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Tyr Tyr Tyr Val Ser Ser Tyr Phe Asp Val Trp Gly Thr Gly
100 105 110

Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala Pro Ser Val Phe
115 120 125

Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser Thr Val Ala Leu
130 135 140

Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val Thr Val Ser Trp
145 150 155 160

Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe Pro Ser Val Leu
165 170 175

Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val Thr Val Pro Ser
180 185 190

Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val Ala His Pro Ala
195 200 205

Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg Glu Asn Gly Arg
210 215 220

Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala Pro Glu Met Leu
225 230 235 240

Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Thr Leu
245 250 255

Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Ala Leu Asp
260 265 270

Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys Gln
275 280 285

Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Ala Gly Thr
290 295 300

Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu Lys
305 310 315 320

Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser Pro
325 330 335

Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro Ser
340 345 350

Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr Val
355 360 365

Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp Val
370 375 380

Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg Thr
385 390 395 400

Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser Lys
405 410 415

Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile Cys
420 425 430

Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser Leu
435 440 445

Ser His Ser Pro Gly Lys Arg Asn Asp Ala Gln Pro Ala Val Tyr Leu
450 455 460

Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly Ser Ala Ser Val Val
465 470 475 480

Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile Asn Val Lys Trp Lys
485 490 495

Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln Glu Ser Val Thr Glu
500 505 510

Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser Thr Leu Thr Met Ser
515 520 525

Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser Cys Glu Ile Thr His
530 535 540

Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe Gln Arg Ser Glu Cys
545 550 555 560

Gln Arg Val Asp

<210> 76
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 76

Glu Ile Val Met Thr Gln Ser Pro Ala Ser Leu Ser Leu Ser Gln Glu
1 5 10 15

Glu Lys Val Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Thr Asn Asp
20 25 30

Val Thr Trp Tyr Gln Gln Lys Pro Gly Gln Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Ser Ala Ser Asn Arg Tyr Thr Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Ser Phe Thr Ile Ser Ser Leu Glu Pro
65 70 75 80

Glu Asp Val Ala Val Tyr Tyr Cys Gln Gln Asp Tyr Ser Ser Pro Trp
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 77
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 77

Asp Ile Val Met Thr Gln Thr Pro Leu Ser Leu Ser Val Ser Pro Gly
1 5 10 15

Glu Thr Ala Ser Ile Ser Cys Lys Ala Ser Gln Ser Val Thr Asn Asp
20 25 30

Val Thr Trp Phe Arg Gln Lys Pro Gly Gln Ser Pro Gln Leu Leu Ile
35 40 45

Tyr Ser Ala Ser Asn Arg Tyr Thr Gly Val Pro Asp Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Arg Ile Ser Thr Val Glu Ala
65 70 75 80

Asp Asp Thr Gly Val Tyr Tyr Cys Gln Gln Asp Tyr Ser Ser Pro Trp
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 78
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 78

Asp Ile Val Met Thr Gln Thr Pro Leu Ser Leu Ser Val Ser Pro Gly
1 5 10 15

Glu Thr Ala Ser Ile Ser Cys Lys Ala Ser Gln Ser Val Thr Asn Asp
20 25 30

Val Thr Trp Phe Arg Gln Lys Pro Gly Gln Ser Pro Lys Leu Leu Ile
35 40 45

Tyr Ser Ala Ser Asn Arg Tyr Thr Gly Val Pro Asp Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Arg Ile Ser Thr Val Glu Ala
65 70 75 80

Asp Asp Thr Gly Val Tyr Tyr Cys Gln Gln Asp Tyr Ser Ser Pro Trp
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 79
<211> 458
<212> PRT
<213> Artificial Sequence

<220>

<223> caninized mouse

<400> 79

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Thr Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Met
35 40 45

Gly His Ile Asn Pro Asn Asn Gly Gly Ile Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Arg Val Thr Leu Thr Ala Asp Thr Ser Thr Ser Thr Ala Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Trp Ala Pro Leu Val Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala
115 120 125

Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser
130 135 140

Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val
145 150 155 160

Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe
165 170 175

Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val
180 185 190

Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val
195 200 205

Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg
210 215 220

Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala
225 230 235 240

Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro
245 250 255

Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val
260 265 270

Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val
275 280 285

Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln
290 295 300

Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln
305 310 315 320

Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala
325 330 335

Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala
340 345 350

His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser
355 360 365

Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro
370 375 380

Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser
385 390 395 400

Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe
405 410 415

Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp
420 425 430

Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr
435 440 445

Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 80
<211> 458
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 80

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Asn Gly Gly Ile Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Ala Asp Thr Ser Thr Ser Thr Ala Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Trp Ala Pro Leu Val Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala
115 120 125

Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser
130 135 140

Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val
145 150 155 160

Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe
165 170 175

Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val
180 185 190

Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val
195 200 205

Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg
210 215 220

Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala
225 230 235 240

Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro
245 250 255

Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val
260 265 270

Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val
275 280 285

Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln
290 295 300

Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln
305 310 315 320

Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala
325 330 335

Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala
340 345 350

His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser
355 360 365

Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro
370 375 380

Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser
385 390 395 400

Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe
405 410 415

Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp
420 425 430

Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr
435 440 445

Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 81
<211> 458
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 81

Glu Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Asp Tyr
20 25 30

Asn Met Asp Trp Val Arg Gln Ala Pro Gly Ala Gly Leu Asp Trp Ile
35 40 45

Gly His Ile Asn Pro Asn Asn Gly Gly Ile Leu Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Val Asp Arg Ser Thr Asn Thr Ala Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ala Gly Asp Ile Ala Val Tyr Phe Cys
85 90 95

Ala Arg Trp Ala Pro Leu Val Arg Gln Pro Tyr Trp Tyr Phe Asp Val
100 105 110

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr Ala
115 120 125

Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly Ser
130 135 140

Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro Val
145 150 155 160

Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr Phe
165 170 175

Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met Val
180 185 190

Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn Val
195 200 205

Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys Arg
210 215 220

Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro Ala
225 230 235 240

Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro
245 250 255

Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val
260 265 270

Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val
275 280 285

Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln
290 295 300

Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln
305 310 315 320

Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala
325 330 335

Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala
340 345 350

His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser
355 360 365

Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro
370 375 380

Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser
385 390 395 400

Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe
405 410 415

Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp
420 425 430

Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr
435 440 445

Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 82
<211> 217
<212> PRT
<213> Artificial Sequence

<220>

<223> caninized mouse

<400> 82

Glu Ile Val Met Thr Gln Ser Pro Ala Ser Leu Ser Leu Ser Gln Glu
1 5 10 15

Glu Lys Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Tyr Gln Gln Lys Pro Gly Gln Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Ser Phe Thr Ile Ser Ser Leu Glu Pro
65 70 75 80

Glu Asp Val Ala Val Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Val Glu Leu Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 83
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 83

Glu Ile Val Met Thr Gln Ser Pro Ala Ser Leu Ser Leu Ser Gln Glu
1 5 10 15

Glu Lys Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Gln Gln Lys Pro Gly Gln Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Ser Phe Thr Ile Ser Ser Leu Glu Pro
65 70 75 80

Glu Asp Val Ala Val Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 84
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 84

Asp Ile Lys Met Thr Gln Ser Pro Ala Ser Leu Ser Leu Ser Gln Glu
1 5 10 15

Glu Lys Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Gln Gln Lys Pro Gly Gln Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Ser Phe Thr Ile Ser Ser Leu Glu Pro
65 70 75 80

Glu Asp Val Ala Val Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 85
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 85

Asp Ile Val Met Thr Gln Thr Pro Leu Ser Leu Ser Val Ser Pro Gly
1 5 10 15

Glu Thr Ala Ser Ile Ser Cys Lys Ala Ser Gln Asp Ile Asn Ser Tyr
20 25 30

Leu Asn Trp Phe Arg Gln Lys Pro Gly Gln Ser Pro Gln Leu Leu Ile
35 40 45

Tyr Arg Ala Asp Arg Leu Val Asp Gly Val Pro Asp Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Arg Ile Ser Thr Val Glu Ala
65 70 75 80

Asp Asp Thr Gly Val Tyr Tyr Cys Leu Gln Tyr Asp Glu Phe Pro Leu
85 90 95

Thr Phe Gly Ala Gly Thr Lys Val Glu Leu Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 86
<211> 459
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 86

Glu Leu Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Val Val Ser Gly Gly Ser Val Thr Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Ile Ser Ile Thr Ala Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr
115 120 125

Ala Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly
130 135 140

Ser Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro
145 150 155 160

Val Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr
165 170 175

Phe Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met
180 185 190

Val Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn
195 200 205

Val Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys
210 215 220

Arg Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro
225 230 235 240

Ala Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys
245 250 255

Pro Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val
260 265 270

Val Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe
275 280 285

Val Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu
290 295 300

Gln Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His
305 310 315 320

Gln Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys
325 330 335

Ala Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln
340 345 350

Ala His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu
355 360 365

Ser Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro
370 375 380

Pro Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu
385 390 395 400

Ser Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr
405 410 415

Phe Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly
420 425 430

Asp Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr
435 440 445

Thr Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 87
<211> 459
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 87

Glu Leu Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Ile Ser Ile Thr Ala Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr
115 120 125

Ala Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly
130 135 140

Ser Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro
145 150 155 160

Val Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr
165 170 175

Phe Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met
180 185 190

Val Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn
195 200 205

Val Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys
210 215 220

Arg Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro
225 230 235 240

Ala Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys
245 250 255

Pro Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val
260 265 270

Val Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe
275 280 285

Val Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu
290 295 300

Gln Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His
305 310 315 320

Gln Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys
325 330 335

Ala Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln
340 345 350

Ala His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu
355 360 365

Ser Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro
370 375 380

Pro Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu
385 390 395 400

Ser Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr
405 410 415

Phe Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly
420 425 430

Asp Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr
435 440 445

Thr Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 88
<211> 459
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 88

Glu Val Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Leu Ser Ile Thr Lys Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr
115 120 125

Ala Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly
130 135 140

Ser Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro
145 150 155 160

Val Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr
165 170 175

Phe Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met
180 185 190

Val Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn
195 200 205

Val Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys
210 215 220

Arg Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro
225 230 235 240

Ala Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys
245 250 255

Pro Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val
260 265 270

Val Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe
275 280 285

Val Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu
290 295 300

Gln Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His
305 310 315 320

Gln Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys
325 330 335

Ala Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln
340 345 350

Ala His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu
355 360 365

Ser Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro
370 375 380

Pro Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu
385 390 395 400

Ser Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr
405 410 415

Phe Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly
420 425 430

Asp Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr
435 440 445

Thr Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 89
<211> 459
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 89

Glu Val Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Leu Ser Ile Thr Lys Asp Thr Ala Lys Asn Gln Val
65 70 75 80

Phe Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Thr
115 120 125

Ala Pro Ser Val Phe Pro Leu Ala Pro Ser Cys Gly Ser Thr Ser Gly
130 135 140

Ser Thr Val Ala Leu Ala Cys Leu Val Ser Gly Tyr Phe Pro Glu Pro
145 150 155 160

Val Thr Val Ser Trp Asn Ser Gly Ser Leu Thr Ser Gly Val His Thr
165 170 175

Phe Pro Ser Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Met
180 185 190

Val Thr Val Pro Ser Ser Arg Trp Pro Ser Glu Thr Phe Thr Cys Asn
195 200 205

Val Ala His Pro Ala Ser Lys Thr Lys Val Asp Lys Pro Val Pro Lys
210 215 220

Arg Glu Asn Gly Arg Val Pro Arg Pro Pro Asp Cys Pro Lys Cys Pro
225 230 235 240

Ala Pro Glu Met Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys
245 250 255

Pro Lys Asp Thr Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val
260 265 270

Val Val Ala Leu Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe
275 280 285

Val Asp Gly Lys Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu
290 295 300

Gln Phe Ala Gly Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His
305 310 315 320

Gln Asp Trp Leu Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys
325 330 335

Ala Leu Pro Ser Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln
340 345 350

Ala His Gln Pro Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu
355 360 365

Ser Lys Asn Thr Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro
370 375 380

Pro Asp Ile Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu
385 390 395 400

Ser Lys Tyr Arg Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr
405 410 415

Phe Leu Tyr Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly
420 425 430

Asp Thr Phe Ile Cys Ala Val Met His Glu Ala Leu His Asn His Tyr
435 440 445

Thr Gln Glu Ser Leu Ser His Ser Pro Gly Lys
450 455

<210> 90
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 90

Glu Ile Val Met Thr Gln Ser Pro Ala Ser Leu Ser Leu Ser Gln Glu
1 5 10 15

Glu Lys Val Thr Ile Thr Cys Arg Ala Ser Glu Asn Ile Tyr Ser Ser
20 25 30

Leu Ala Trp Tyr Gln Gln Lys Pro Gly Gln Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Ala Ala Thr Asn Leu Ala Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Ser Phe Thr Ile Ser Ser Leu Glu Pro
65 70 75 80

Glu Asp Val Ala Val Tyr Tyr Cys Gln His Phe Arg Asp Thr Pro Pro
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 91
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 91

Asp Ile Val Met Thr Gln Thr Pro Leu Ser Leu Ser Val Ser Pro Gly
1 5 10 15

Glu Thr Ala Ser Ile Ser Cys Arg Ala Ser Glu Asn Ile Tyr Ser Ser
20 25 30

Leu Ala Trp Phe Arg Gln Lys Pro Gly Gln Ser Pro Gln Leu Leu Ile
35 40 45

Tyr Ala Ala Thr Asn Leu Ala Asp Gly Val Pro Asp Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Arg Ile Ser Arg Val Glu Ala
65 70 75 80

Asp Asp Thr Gly Val Tyr Tyr Cys Gln His Phe Arg Asp Thr Pro Pro
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 92
<211> 217
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 92

Asp Ile Val Met Thr Gln Thr Pro Leu Ser Leu Ser Val Ser Pro Gly
1 5 10 15

Glu Thr Ala Ser Ile Ser Cys Arg Ala Ser Glu Asn Ile Tyr Ser Ser
20 25 30

Leu Ala Trp Phe Arg Gln Lys Pro Gly Gln Ser Pro Gln Leu Leu Val
35 40 45

Tyr Ala Ala Thr Asn Leu Ala Asp Gly Val Pro Asp Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Tyr Thr Leu Arg Ile Ser Arg Val Glu Ala
65 70 75 80

Asp Asp Thr Gly Val Tyr Tyr Cys Gln His Phe Arg Asp Thr Pro Pro
85 90 95

Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg Asn Asp Ala Gln
100 105 110

Pro Ala Val Tyr Leu Phe Gln Pro Ser Pro Asp Gln Leu His Thr Gly
115 120 125

Ser Ala Ser Val Val Cys Leu Leu Asn Ser Phe Tyr Pro Lys Asp Ile
130 135 140

Asn Val Lys Trp Lys Val Asp Gly Val Ile Gln Asp Thr Gly Ile Gln
145 150 155 160

Glu Ser Val Thr Glu Gln Asp Lys Asp Ser Thr Tyr Ser Leu Ser Ser
165 170 175

Thr Leu Thr Met Ser Ser Thr Glu Tyr Leu Ser His Glu Leu Tyr Ser
180 185 190

Cys Glu Ile Thr His Lys Ser Leu Pro Ser Thr Leu Ile Lys Ser Phe
195 200 205

Gln Arg Ser Glu Cys Gln Arg Val Asp
210 215

<210> 93
<211> 124
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 93

Glu Leu Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Val Val Ser Gly Gly Ser Val Thr Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Ile Ser Ile Thr Ala Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
115 120

<210> 94
<211> 124
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 94

Glu Leu Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Ile Ser Ile Thr Ala Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
115 120

<210> 95
<211> 124
<212> PRT
<213> Artificial Sequence

<220>
<223> caninized mouse

<400> 95

Glu Val Thr Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Leu Ser Ile Thr Lys Asp Thr Ala Lys Asn Gln Phe
65 70 75 80

Ser Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
115 120

<210> 96
<211> 124
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<400> 96

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1 5 10 15

Thr Leu Ser Leu Thr Cys Ser Phe Ser Gly Phe Ser Leu Ser Thr Phe
20 25 30

Gly Arg Gly Val Gly Trp Ile Arg Gln Arg Pro Gly Arg Gly Leu Glu
35 40 45

Trp Met Gly His Ile Trp Trp Asp Asp Asp Lys Tyr Tyr Asn Pro Ala
50 55 60

Leu Lys Ser Arg Leu Ser Ile Thr Lys Asp Thr Ala Lys Asn Gln Val
65 70 75 80

Phe Leu Gln Leu Ser Ser Met Thr Thr Glu Asp Thr Ala Val Tyr Tyr
85 90 95

Cys Ala Arg Ile Ala Gly Gly Leu Arg Arg Ala Pro Tyr Ala Met Asp
100 105 110

Ser Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
115 120

<210> 97
<211> 19
<212> PRT
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<400> 97

Tyr Trp Asn Leu Asp Ala Ile Met Lys Ile Glu Pro Pro Glu Ile Phe
1 5 10 15

Ser Val Lys

<210> 98
<211> 18
<212> PRT
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<400> 98

Lys Ile Glu Pro Pro Glu Ile Phe Ser Val Lys Ser Val Leu Gly Ile
1 5 10 15

Lys Arg

<210> 99
<211> 13
<212> PRT
<213> Canis familiaris

<400> 99

Arg Pro Val Leu Ala Pro His Ser Ser Thr Leu Lys Tyr
1 5 10

<210> 100
<211> 14
<212> PRT
<213> Canis familiaris

<400> 100

Thr Glu Tyr Val Met Thr Leu Arg Cys Ala Pro Ala Glu Ser
1 5 10

<210> 101
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<212> PRT
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<400> 101

Tyr Val Met Thr Leu Arg

1 5

<210> 102
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<212> PRT
<213> Canis familiaris

<400> 102

Arg Cys Ala Pro Ala Glu Ser Met
1 5

<210> 103
<211> 20
<212> PRT
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<400> 103

Arg Cys Ala Pro Ala Glu Ser Met Phe Trp Ser Gly Trp Ser Gln Glu
1 5 10 15

Lys Val Gly Thr
20

<210> 104
<211> 12
<212> PRT
<213> Canis familiaris

<400> 104

Arg Arg Pro Val Gln Leu Met Trp Lys Lys Ala Thr
1 5 10

<210> 105
<211> 9
<212> PRT
<213> Canis familiaris

<400> 105

Arg Ile Pro Ala Leu Asn Glu Lys Thr
1 5

<210> 106
<211> 12
<212> PRT

<213> Canis familiaris

<400> 106

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1 5 10

<210> 107
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<400> 107

Arg Thr Ser Tyr Ser Leu Gln Val Met Ala Ser Thr Asn Ala Gly Gly
1 5 10 15

Thr

<210> 108
<211> 16
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<400> 108

Ser Leu Gln Val Met Ala Ser Thr Asn Ala Gly Gly Thr Asn Gly Thr
1 5 10 15

<210> 109
<211> 10
<212> PRT
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<400> 109

Ser Thr Asn Ala Gly Gly Thr Asn Gly Thr
1 5 10

<210> 110
<211> 215
<212> PRT
<213> Canis familiaris

<400> 110

Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Thr
1 5 10 15

Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Asp Leu
20 25 30

Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys
35 40 45

Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Asn Gly
50 55 60

Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu
65 70 75 80

Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser
85 90 95

Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro
100 105 110

Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr
115 120 125

Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp
130 135 140

Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg
145 150 155 160

Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser
165 170 175

Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile
180 185 190

Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser
195 200 205

Leu Ser His Ser Pro Gly Lys
210 215

<210> 111
<211> 214
<212> PRT
<213> Artificial Sequence

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<223> modified canine

<400> 111

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Leu Leu Ile Ala Arg Thr Pro Glu Val Thr Cys Val Val Val Ala Leu
20 25 30

Asp Pro Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys
35 40 45

Gln Met Gln Thr Ala Lys Thr Gln Pro Arg Glu Glu Gln Phe Ala Gly
50 55 60

Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu
65 70 75 80

Lys Gly Lys Gln Phe Thr Cys Lys Val Asn Asn Lys Ala Leu Pro Ser
85 90 95

Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro
100 105 110

Ser Val Tyr Val Leu Pro Pro Ser Arg Glu Glu Leu Ser Lys Asn Thr
115 120 125

Val Ser Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Asp Ile Asp
130 135 140

Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg
145 150 155 160

Thr Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser
165 170 175

Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile
180 185 190

Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Glu Ser
195 200 205

Leu Ser His Ser Pro Gly
210

<210> 112
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<212> PRT
<213> Canis familiaris

<400> 112

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1 5 10 15

Pro

<210> 113
<211> 22
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<400> 113

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1 5 10 15

Cys Pro Ala Pro Glu Met
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<210> 114
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Ala Lys Glu Cys Glu Cys Lys Cys Asn Cys Asn Asn Cys Pro Cys Pro
1 5 10 15

Gly Cys Gly Leu

20

<210> 115
<211> 17
<212> PRT
<213> Artificial Sequence

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<400> 115

Pro Lys Glu Ser Thr Cys Lys Cys Ile Pro Pro Cys Pro Val Pro Glu
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Ser

<210> 116
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<213> Canis familiaris

<400> 116

Leu Gly Gly Pro Ser Val Leu Ile Phe Pro Pro Lys Pro Lys Asp Ile
1 5 10 15

Leu Arg Ile Thr Arg Thr Pro Glu Val Thr Cys Val Val Leu Asp Leu
20 25 30

Gly Arg Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys
35 40 45

Glu Val His Thr Ala Lys Thr Gln Ser Arg Glu Gln Gln Phe Asn Gly
50 55 60

Thr Tyr Arg Val Val Ser Val Leu Pro Ile Glu His Gln Asp Trp Leu
65 70 75 80

Thr Gly Lys Glu Phe Lys Cys Arg Val Asn His Ile Asp Leu Pro Ser
85 90 95

Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Arg Ala His Lys Pro
100 105 110

Ser Val Tyr Val Leu Pro Pro Ser Pro Lys Glu Leu Ser Ser Ser Asp
115 120 125

Thr Val Ser Ile Thr Cys Leu Ile Lys Asp Phe Tyr Pro Pro Asp Ile
130 135 140

Asp Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Arg Lys His
145 150 155 160

Arg Met Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr
165 170 175

Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Gln Gly Asp Pro Phe
180 185 190

Thr Cys Ala Val Met His Glu Thr Leu Gln Asn His Tyr Thr Asp Leu
195 200 205

Ser Leu Ser His Ser Pro Gly Lys
210 215

<210> 117
<211> 215
<212> PRT
<213> Canis familiaris

<400> 117

Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Ile
1 5 10 15

Leu Val Thr Ala Arg Thr Pro Thr Val Thr Cys Val Val Val Asp Leu
20 25 30

Asp Pro Glu Asn Pro Glu Val Gln Ile Ser Trp Phe Val Asp Ser Lys
35 40 45

Gln Val Gln Thr Ala Asn Thr Gln Pro Arg Glu Glu Gln Ser Asn Gly
50 55 60

Thr Tyr Arg Val Val Ser Val Leu Pro Ile Gly His Gln Asp Trp Leu

65 70 75 80

Ser Gly Lys Gln Phe Lys Cys Lys Val Asn Asn Lys Ala Leu Pro Ser
85 90 95

Pro Ile Glu Glu Ile Ile Ser Lys Thr Pro Gly Gln Ala His Gln Pro
100 105 110

Asn Val Tyr Val Leu Pro Pro Ser Arg Asp Glu Met Ser Lys Asn Thr
115 120 125

Val Thr Leu Thr Cys Leu Val Lys Asp Phe Phe Pro Pro Glu Ile Asp
130 135 140

Val Glu Trp Gln Ser Asn Gly Gln Gln Glu Pro Glu Ser Lys Tyr Arg
145 150 155 160

Met Thr Pro Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr Ser
165 170 175

Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Arg Gly Asp Thr Phe Ile
180 185 190

Cys Ala Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Ile Ser
195 200 205

Leu Ser His Ser Pro Gly Lys
210 215

<210> 118
<211> 216
<212> PRT
<213> Canis familiaris

<400> 118

Leu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Lys Pro Lys Asp Ile
1 5 10 15

Leu Arg Ile Thr Arg Thr Pro Glu Ile Thr Cys Val Val Leu Asp Leu
20 25 30

Gly Arg Glu Asp Pro Glu Val Gln Ile Ser Trp Phe Val Asp Gly Lys
35 40 45

Glu Val His Thr Ala Lys Thr Gln Pro Arg Glu Gln Gln Phe Asn Ser
50 55 60

Thr Tyr Arg Val Val Ser Val Leu Pro Ile Glu His Gln Asp Trp Leu
65 70 75 80

Thr Gly Lys Glu Phe Lys Cys Arg Val Asn His Ile Gly Leu Pro Ser
85 90 95

Pro Ile Glu Arg Thr Ile Ser Lys Ala Arg Gly Gln Ala His Gln Pro
100 105 110

Ser Val Tyr Val Leu Pro Pro Ser Pro Lys Glu Leu Ser Ser Ser Asp
115 120 125

Thr Val Thr Leu Thr Cys Leu Ile Lys Asp Phe Phe Pro Pro Glu Ile
130 135 140

Asp Val Glu Trp Gln Ser Asn Gly Gln Pro Glu Pro Glu Ser Lys Tyr
145 150 155 160

His Thr Thr Ala Pro Gln Leu Asp Glu Asp Gly Ser Tyr Phe Leu Tyr
165 170 175

Ser Lys Leu Ser Val Asp Lys Ser Arg Trp Gln Gln Gly Asp Thr Phe
180 185 190

Thr Cys Ala Val Met His Glu Ala Leu Gln Asn His Tyr Thr Asp Leu
195 200 205

Ser Leu Ser His Ser Pro Gly Lys
210 215

<210> 119
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<212> PRT
<213> Canis familiaris

<400> 119

Ser Asp Ile Thr Tyr Trp Asn Leu Asp Ala Ile Met Lys Ile Glu Pro
1 5 10 15

Pro Glu Ile Phe Ser Val Lys Ser Val Leu Gly Ile Lys Arg Met
20 25 30

<210> 120
<211> 31
<212> PRT
<213> Canis familiaris

<400> 120

Met Thr Leu Arg Cys Ala Pro Ala Glu Ser Met Phe Trp Ser Gly Trp
1 5 10 15

Ser Gln Glu Lys Val Gly Thr Thr Glu Glu Glu Ala Pro Tyr Gly
20 25 30

<210> 121
<211> 21
<212> PRT
<213> Canis familiaris

<400> 121

Ile Met Lys Ile Glu Pro Pro Glu Ile Phe Ser Val Lys Ser Val Leu
1 5 10 15

Gly Ile Lys Arg Met
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<210> 122
<211> 21
<212> PRT
<213> Canis familiaris

<400> 122

Thr Glu Leu Gln Ala Phe Thr Glu Tyr Val Met Thr Leu Arg Cys Ala
1 5 10 15

Pro Ala Glu Ser Met
20

<210> 123
<211> 21
<212> PRT
<213> Canis familiaris

<400> 123

Ala Met Val Asp Gly Arg Arg Pro Val Gln Leu Met Trp Lys Lys Ala
1 5 10 15

Thr Gly Ala Pro Val
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<210> 124
<211> 21
<212> PRT
<213> Canis familiaris

<400> 124

Gly Glu Ser Pro Val Ala Thr Leu Arg Ile Pro Ala Leu Asn Glu Lys
1 5 10 15

Thr Phe Gln Cys Ile
20

<210> 125
<211> 21
<212> PRT
<213> Canis familiaris

<400> 125

Arg Asn Gly Phe Ile Lys Asn Tyr Thr Ile Phe Tyr Gln Ala Glu Asp
1 5 10 15

Gly Lys Glu Phe Ser
20

<210> 126
<211> 21
<212> PRT
<213> Canis familiaris

<400> 126

Ser Tyr Ser Leu Gln Val Met Ala Ser Thr Asn Ala Gly Gly Thr Asn
1 5 10 15

Gly Thr Lys Ile Asn
20

5

WHAT IS CLAIMED IS:

1. A mammalian antibody or antigen binding fragment thereof that binds canine interleukin-31 receptor *alpha* (canine IL-31RA) comprising a heavy chain that comprises a set of three heavy chain complementary determining regions (HCDRs) each comprising an amino acid sequence: a CDR heavy 1 (HCDR1), a CDR heavy 2 (HCDR2), and a CDR heavy 3 (HCDR3); wherein

(i) the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3;

(ii) the HCDR2 comprises the amino acid sequence selected from the group consisting of SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, and SEQ ID NO: 13;

(iii) the HCDR3 comprises the amino acid sequence selected from the group consisting of SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, and SEQ ID NO: 23.

2. The mammalian antibody or antigen binding fragment thereof of Claim 1, that when bound to canine IL-31RA the antibody binds to an epitope comprised by an amino acid sequence selected from the group consisting of SEQ ID NO: 97, SEQ ID NO: 103, SEQ ID NO: 99, SEQ ID NO: 102, or any combination thereof.

3. The mammalian antibody or antigen binding fragment thereof of Claim 2, that when bound to canine IL-31RA the antibody binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 97 and SEQ ID NO: 103.

4. The mammalian antibody or antigen binding fragment thereof of Claim 2, that when bound to canine IL-31RA the antibody binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 98 and SEQ ID NO: 102.

5. The mammalian antibody or antigen binding fragment thereof of Claim 1 or 3, wherein

(i) the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3;

(ii) the HCDR2 comprises the amino acid sequence of SEQ ID NO: 10; and

(iii) the HCDR3 comprises the amino acid sequence of SEQ ID NO: 20.

6. The mammalian antibody or antigen binding fragment thereof of any one of Claims 1-4, wherein

(i) the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3;

(ii) the HCDR2 comprises the amino acid sequence of SEQ ID NO: 11; and

(iii) the HCDR3 comprises the amino acid sequence of SEQ ID NO: 21.

7. The mammalian antibody or antigen binding fragment thereof of any one of Claims 1-4, wherein

(i) the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3;

(ii) the HCDR2 comprises the amino acid sequence of SEQ ID NO: 12; and

(iii) the HCDR3 comprises the amino acid sequence of SEQ ID NO: 22.

8. The mammalian antibody or antigen binding fragment thereof of Claim 1 or 4, wherein

(i) the HCDR1 comprises the amino acid sequence of SEQ ID NO: 3;

(ii) the HCDR2 comprises the amino acid sequence of SEQ ID NO: 13; and

(iii) the HCDR3 comprises the amino acid sequence of SEQ ID NO: 23.

9. The mammalian antibody or antigen binding fragment thereof of any one of Claims 1-8, further comprising a light chain that comprises a set of three light chain complementary determining regions (LCDRs) each comprising an amino acid sequence: a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3); wherein

(iv) the LCDR1 comprises the amino acid sequence selected from the group consisting of SEQ ID NO: 30 and SEQ ID NO: 31;

(v) the LCDR2 comprises the amino acid sequence selected from the group consisting of SEQ ID NO: 38, SEQ ID NO: 39, and SEQ ID NO: 40; and

(vi) the LCDR3 comprises the amino acid sequence of SEQ ID NO: 45.

10. The mammalian antibody or antigen binding fragment thereof of Claim 5 or 6, further comprising a set of three light chain complementary determining regions (LCDRs): a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3); wherein

(iv) the LCDR1 comprises the amino acid sequence of SEQ ID NO: 30;

(v) the LCDR2 comprises the amino acid sequence of SEQ ID NO: 40; and

(vi) the LCDR3 comprises the amino acid sequence of SEQ ID NO: 45.

11. The mammalian antibody or antigen binding fragment thereof of Claim 7, further comprising a set of three light chain complementary determining regions (LCDRs): a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3); wherein

(iv) the LCDR1 comprises the amino acid sequence of SEQ ID NO: 30;

(v) the LCDR2 comprises the amino acid sequence of SEQ ID NO: 38; and

(vi) the LCDR3 comprises the amino acid sequence of SEQ ID NO: 45.

12. The mammalian antibody or antigen binding fragment thereof of Claim 8, further comprising a set of three light chain complementary determining regions (LCDRs): a CDR light 1 (LCDR1), a CDR light 2 (LCDR2), and a CDR light 3 (LCDR3); wherein

(iv) the LCDR1 comprises the amino acid sequence of SEQ ID NO: 31;

(v) the LCDR2 comprises the amino acid sequence of SEQ ID NO: 39; and

(vi) the LCDR3 comprises the amino acid sequence of SEQ ID NO: 45.

13. The mammalian antibody or antigen binding fragment thereof of any one of Claims 1 to 12, wherein the antibody and antigen binding fragment thereof bind canine IL-31RA and block the binding of canine IL-31RA to canine interleukin-31.

14. The mammalian antibody or antigen binding fragment thereof of any one of Claims 1-13, wherein the mammalian antibody or antigen binding fragment thereof is a caninized antibody or a caninized antigen binding fragment thereof.

15. The caninized antibody or antigen binding fragment thereof of Claim 14, that comprises a hinge region that comprises an amino acid sequence that comprises at least 90%, 95%, or 100% identity with the amino acid sequence selected from the group consisting of SEQ ID NO: 112, SEQ ID NO: 113, SEQ ID NO: 114, and SEQ ID NO: 115.

16. The caninized antibody or antigen binding fragment thereof of Claim 14 or 15, that comprises a canine fragment crystallizable region (cFc region); wherein the cFc region comprises an amino acid sequence that comprises at least 90%, 95%, 98%, 99%, or 100%

identity with the amino acid sequence selected from the group consisting of SEQ ID NO: 110, SEQ ID NO: 116, SEQ ID NO: 117, and SEQ ID NO: 118.

17. The caninized antibody or antigen binding fragment thereof of Claim 14 or 15, that comprises a canine fragment crystallizable region (cFc region); wherein the cFc region comprises an amino acid sequence that comprises at least 90%, 95%, 98%, 99%, or 100% identity with the amino acid sequence SEQ ID NO: 111, wherein both the aspartic acid residue (D) at position 31 of SEQ ID NO: 110 and the asparagine residue (N) at position 63 of SEQ ID NO: 110, remain substituted by an alanine residue (A).

18. A caninized antibody or antigen binding fragment thereof, wherein the caninized IL-31RA antibody comprises a light chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO: 82, SEQ ID NO: 83, SEQ ID NO: 84, and SEQ ID NO: 85; and a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO: 79, SEQ ID NO: 80, and SEQ ID NO: 81.

19. The caninized antibody or antigen binding fragment thereof of Claim 18, that when bound to canine IL-31RA the antibody binds to an epitope comprised by an amino acid sequence of SEQ ID NO: 97 and SEQ ID NO: 103.

20. The caninized antibody or antigen binding fragment thereof of Claim 18 or 19, wherein the caninized IL-31RA antibody comprises:

a light chain comprising the amino acid sequence of SEQ ID NO: 84 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80; or

a light chain comprising the amino acid sequence of SEQ ID NO: 84 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81; or

a light chain comprising the amino acid sequence of SEQ ID NO: 85 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 80; or

a light chain comprising the amino acid sequence of SEQ ID NO: 85 and a heavy chain comprising the amino acid sequence of SEQ ID NO: 81.

21. A nucleic acid that encodes the heavy chain of the caninized antibody or antigen binding fragment thereof of any one of Claims 14-20.

22. A nucleic acid that encodes the light chain of the caninized antibody or antigen binding fragment thereof of any one of Claims 14-20.

23. A pair of nucleic acids, wherein one of the pair of nucleic acids comprises a nucleotide sequence that encodes the light chain of a specific caninized antibody of any one of the antibodies of Claims 14-20 and the other of the pair of nucleic acids comprises a nucleotide sequence that encodes the heavy chain of said specific caninized antibody.

24. An expression vector comprising the pair of nucleic acids of Claim 23 or the nucleic acid of Claim 21 or 22.

25. A pair of expression vectors, wherein one of the pair of expression vectors comprises a nucleic acid comprising a nucleotide sequence that encodes the light chain of a specific caninized antibody of any one of the antibodies of Claims 14-20 and the other of the pair of expression vectors comprises a nucleic acid comprising a nucleotide sequence that encodes the heavy chain of said specific caninized antibody.

26. A host cell comprising the expression vector of Claim 24 or the pair of expression vectors of Claim 25.

27. A pharmaceutical composition comprising the caninized antibody or antigen binding fragment thereof of Claims 14-20, and a pharmaceutically acceptable carrier or diluent.

28. A method of aiding in blocking pruritus associated with atopic dermatitis in an animal subject, comprising administering to the animal subject a therapeutically effective amount of the pharmaceutical composition of Claim 27.

29. A pharmaceutical composition for use in aiding in blocking pruritus associated with atopic dermatitis in a canine comprising the caninized antibody or antigen binding fragment thereof of Claims 14-20, and a pharmaceutically acceptable carrier or diluent.

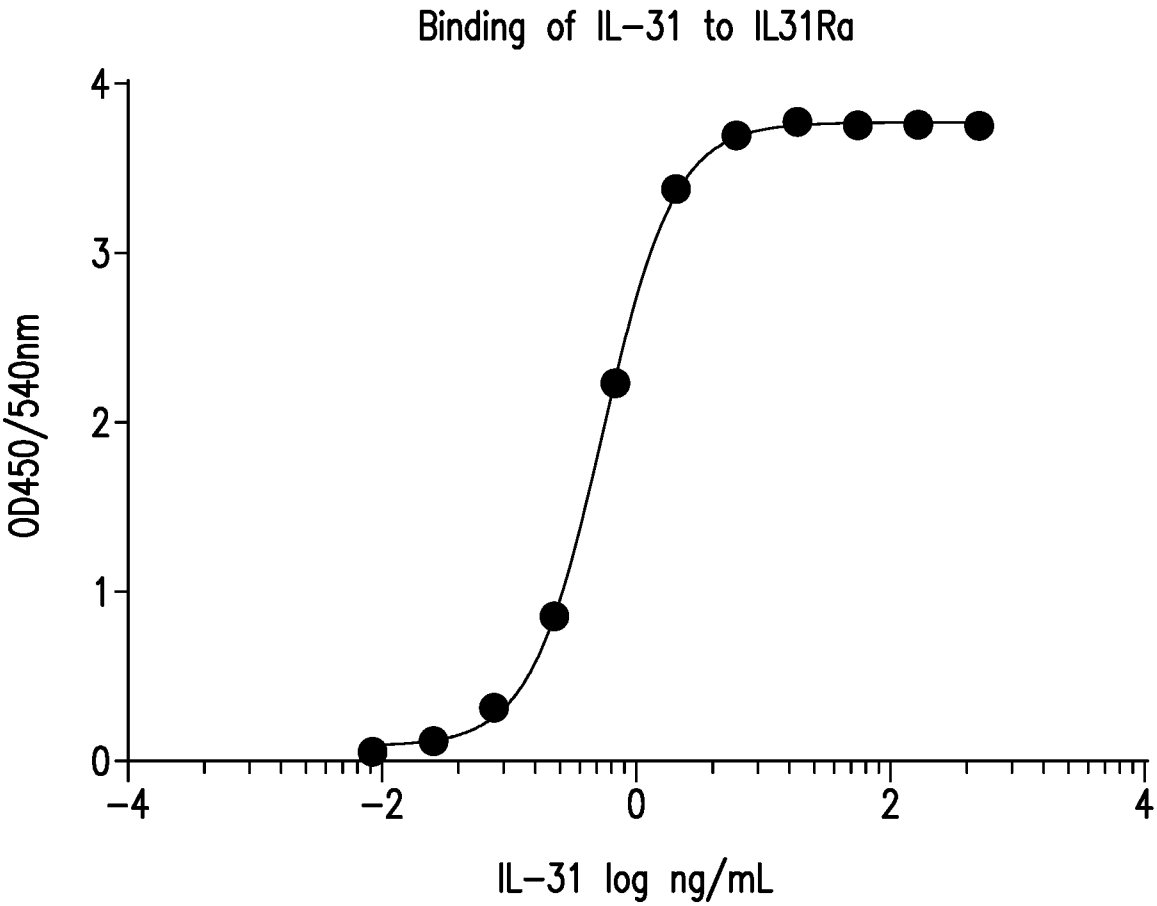

Binding of IL-31 to IL31Ra
OD450/540nm
0
1
2
3
4
−4
−2
0
2
4
IL-31 log ng/mL

FIG.1

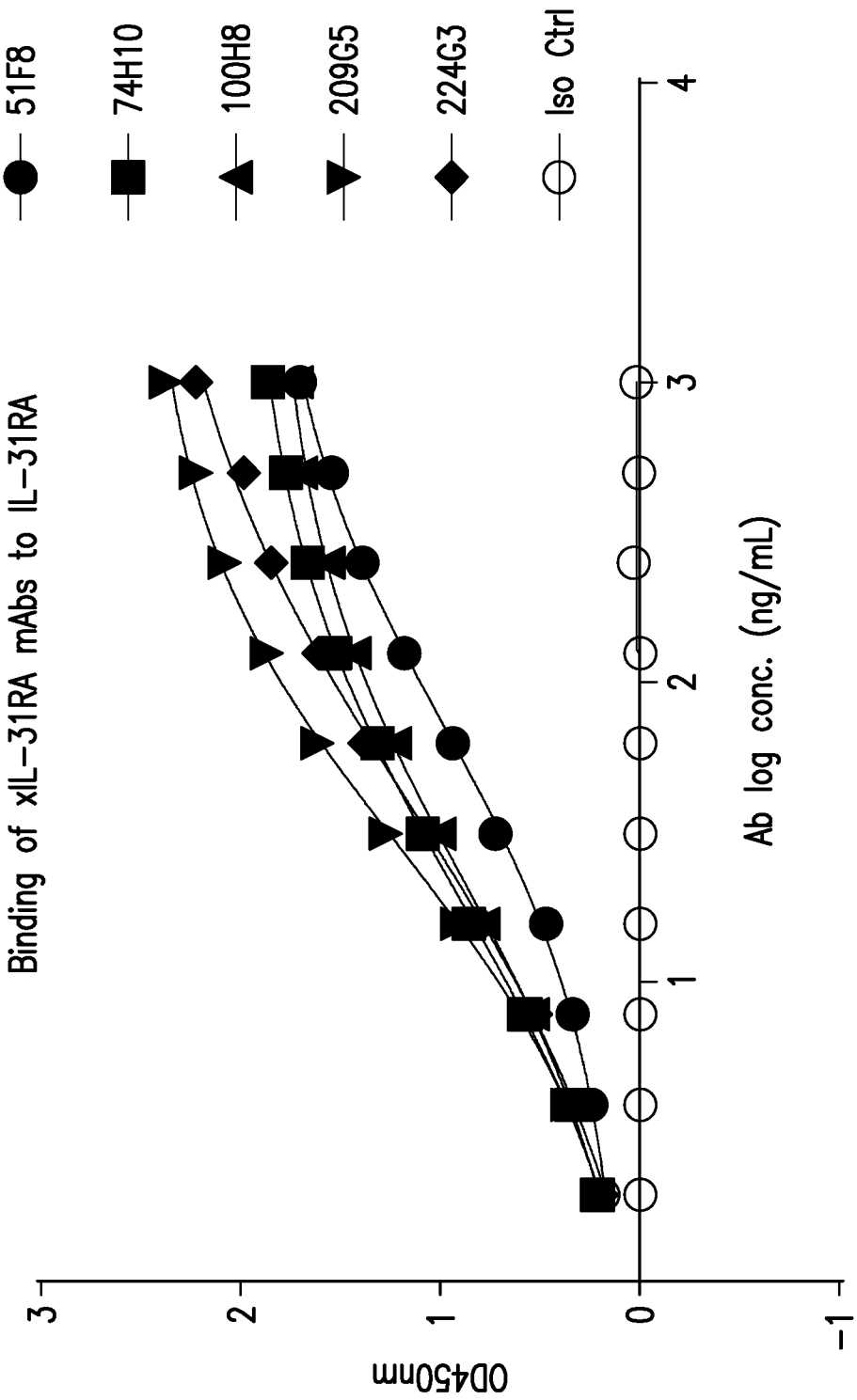

Binding of xIL-31RA mAbs to IL-31RA
51F8
74H10
100H8
209G5
224G3
Iso Ctrl
OD450nm
3
2
1
0
-1
1
2
3
4
Ab log conc. (ng/mL)

FIG.2A

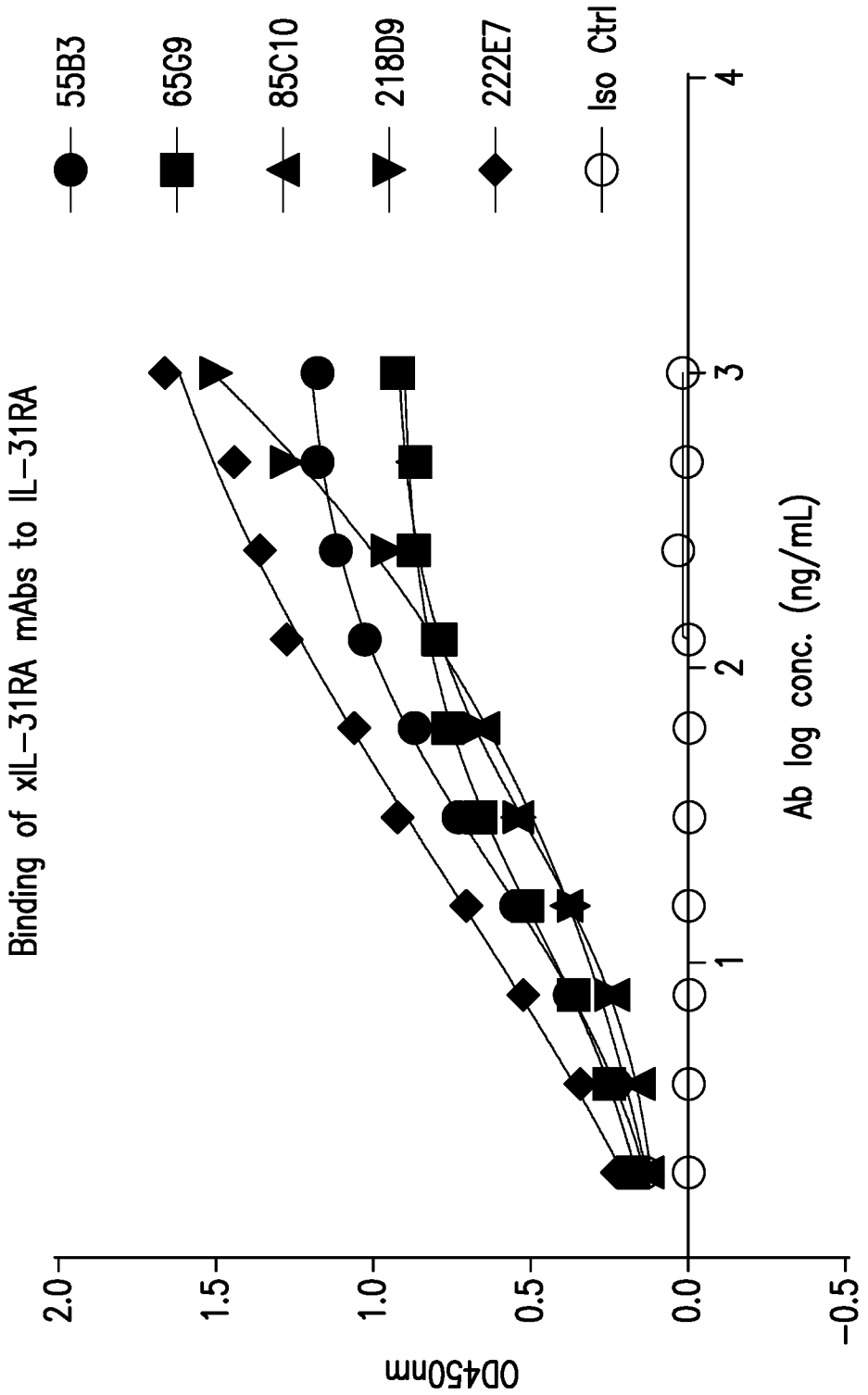

Binding of xIL-31RA mAbs to IL-31RA
55B3
65G9
85C10
218D9
222E7
Iso Ctrl
OD450nm
2.0
1.5
1.0
0.5
0.0
-0.5
1
2
3
4
Ab log conc. (ng/mL)

FIG.2B

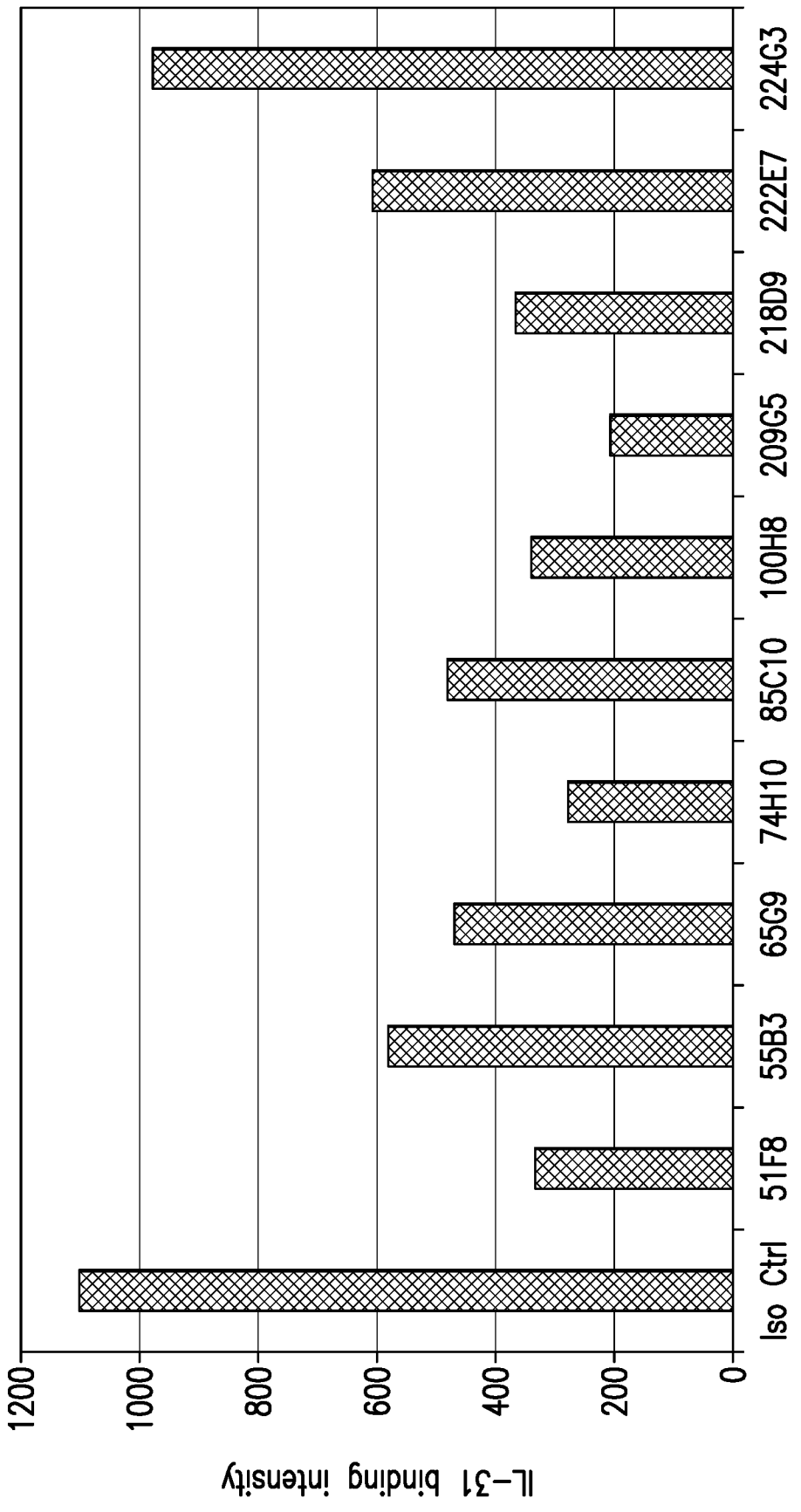
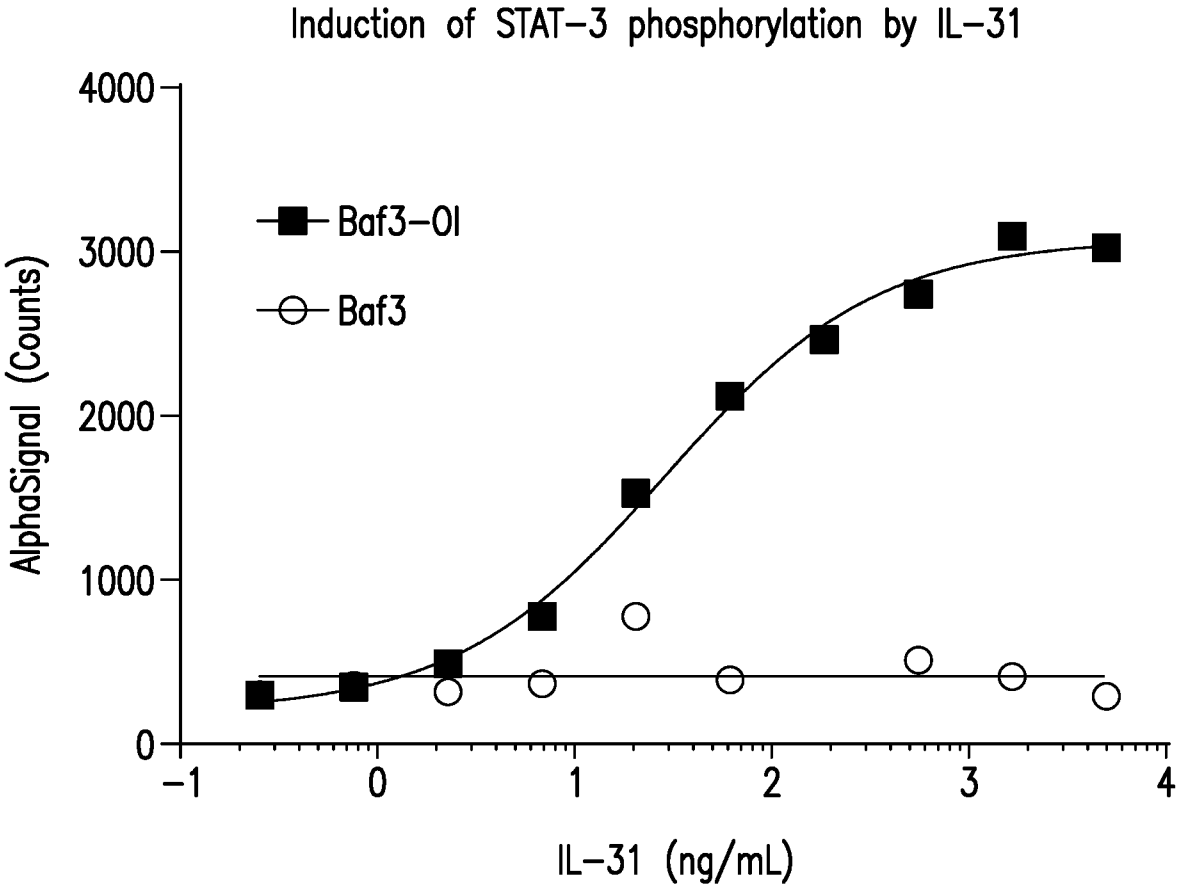

Blocking of IL-31 binding by the selected anti-IL-31RA antibodies
IL-31 binding intensity
0
200
400
600
800
1000
1200
Iso Ctrl
51F8
55B3
65G9
74H10
85C10
100H8
209G5
218D9
222E7
224G3
Anti-IL-31RA Antibody

FIG.3



	Baf3	Baf3-OI
EC50	~ 0.000	29.16

FIG.4

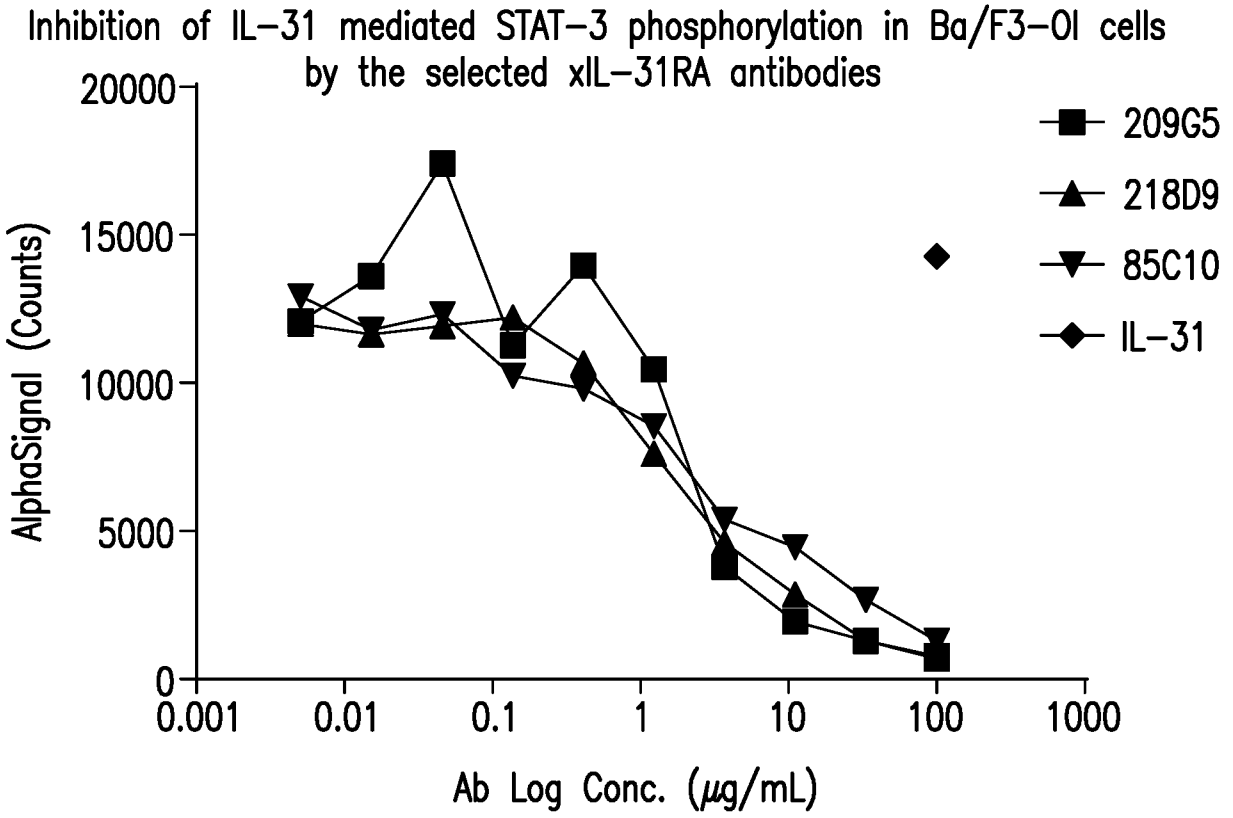

Inhibition of IL-31 mediated STAT-3 phosphorylation in Ba/F3-OI cells
by the selected xIL-31RA antibodies
AlphaSignal (Counts)
20000
15000
10000
5000
0
0.001
0.01
0.1
1
10
100
1000
Ab Log Conc. (µg/mL)
209G5
218D9
85C10
IL-31

FIG.5A

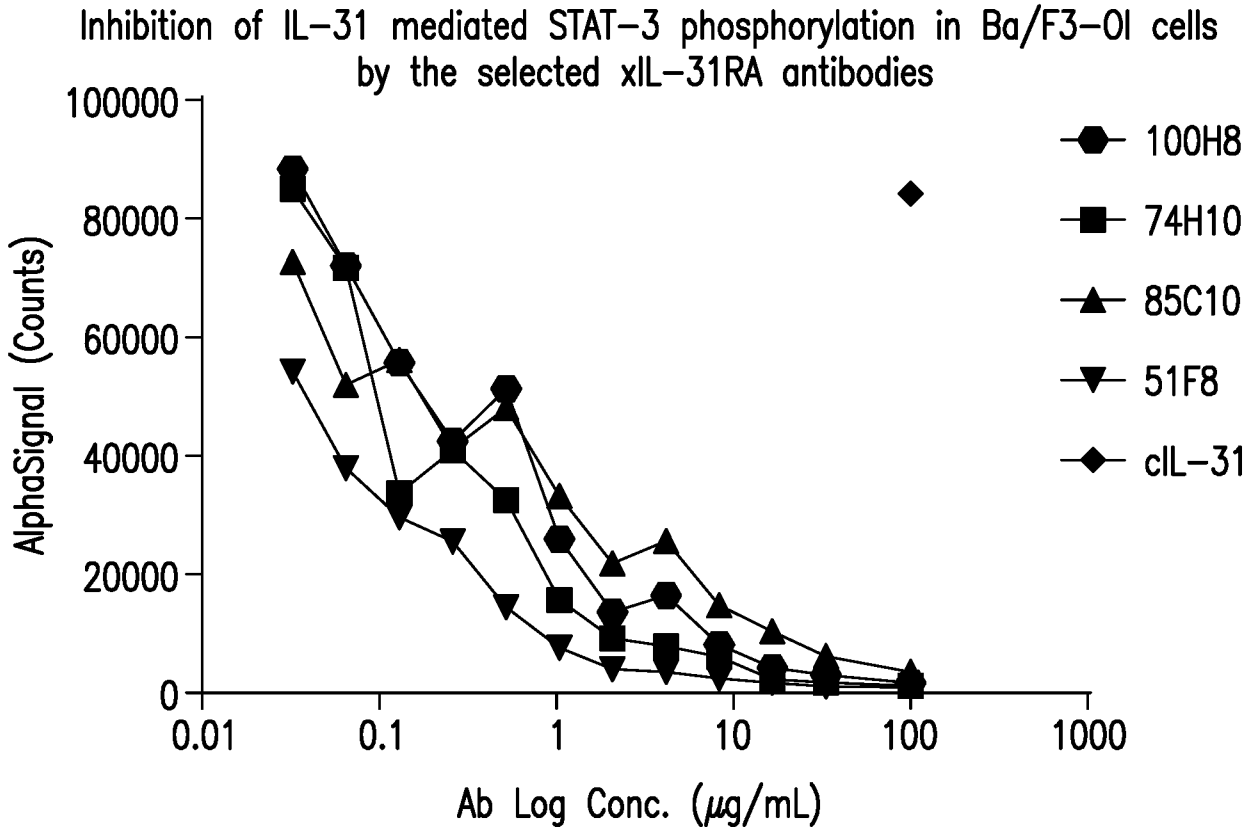

Inhibition of IL-31 mediated STAT-3 phosphorylation in Ba/F3-OI cells
by the selected xIL-31RA antibodies
AlphaSignal (Counts)
100000
80000
60000
40000
20000
0
0.01
0.1
1
10
100
1000
Ab Log Conc. (µg/mL)
100H8
74H10
85C10
51F8
cIL-31

FIG.5B

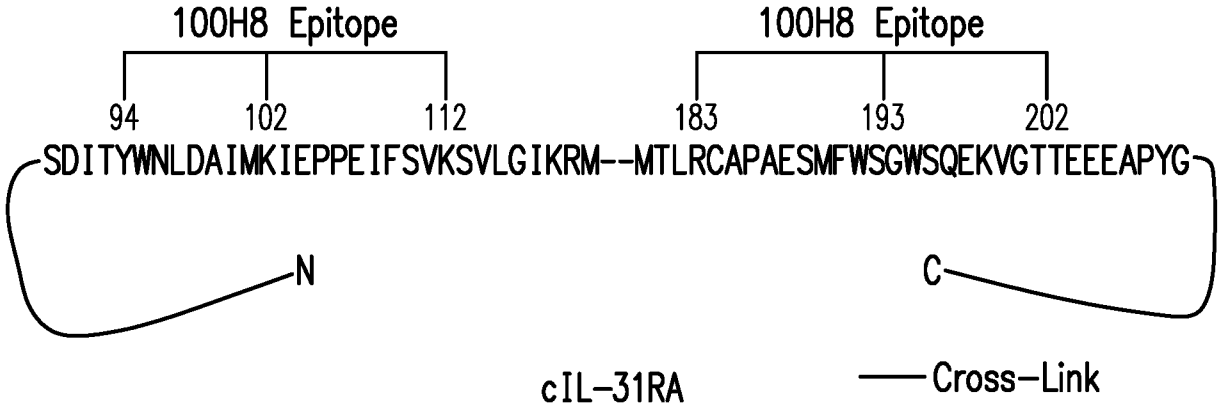

100H8 Epitope
100H8 Epitope
94
102
112
183
193
202
SDITYWNLDAIMKIEPPEIFSVKSVLGIKRM--MTLRCAPAESMFWSGWSQEKVGTTEEEAPYG
N
C
cIL-31RA
Cross-Link

FIG.6A

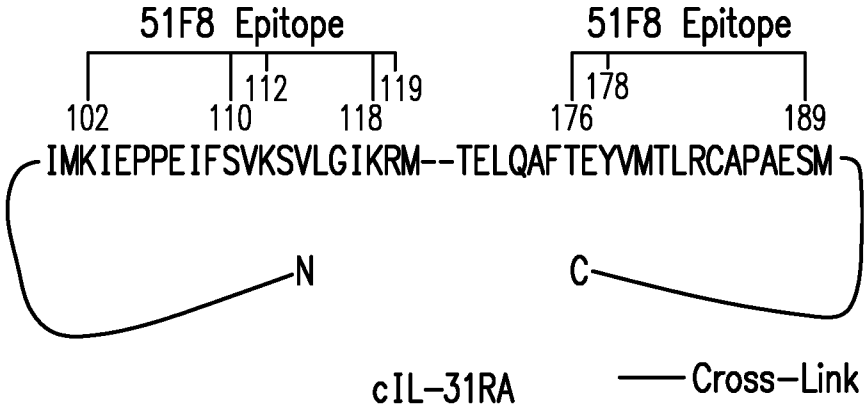

51F8 Epitope
51F8 Epitope
102
110
112
118
119
176
178
189
IMKIEPPEIFSVKSVLGIKRM--TELQAFTEYVMTLRCAPAESM
N
C
cIL-31RA
Cross-Link

FIG.6B

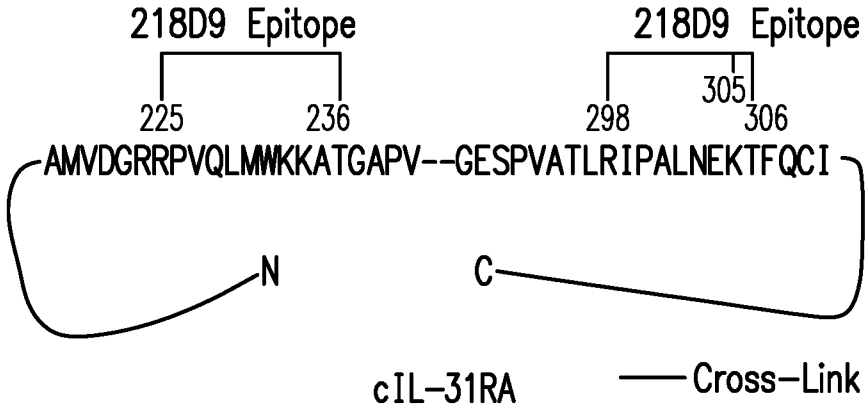

218D9 Epitope
218D9 Epitope
225
236
298
305
306
AMVDGRRPVQLMWKKATGAPV--GESPVATLRIPALNEKTFQCI
N
C
cIL-31RA
Cross-Link

FIG.6C

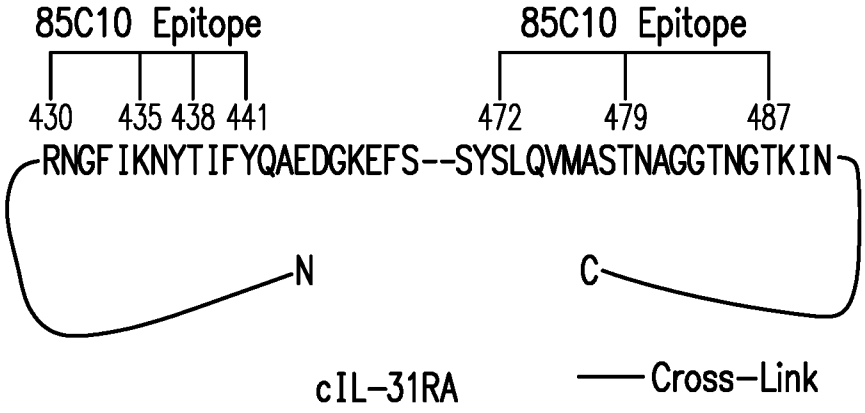

85C10 Epitope
85C10 Epitope
430
435
438
441
472
479
487
RNGFIKNYTIFYQAEDGKEFS--SYSLQVMASTNAGGTNGTKIN
N
C
cIL-31RA
Cross-Link

FIG.6D

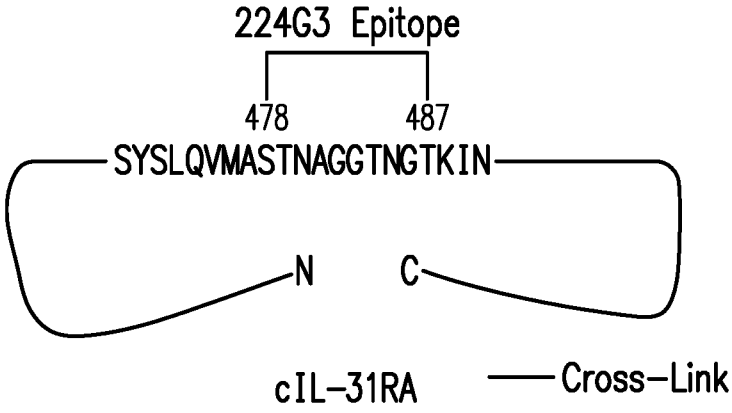
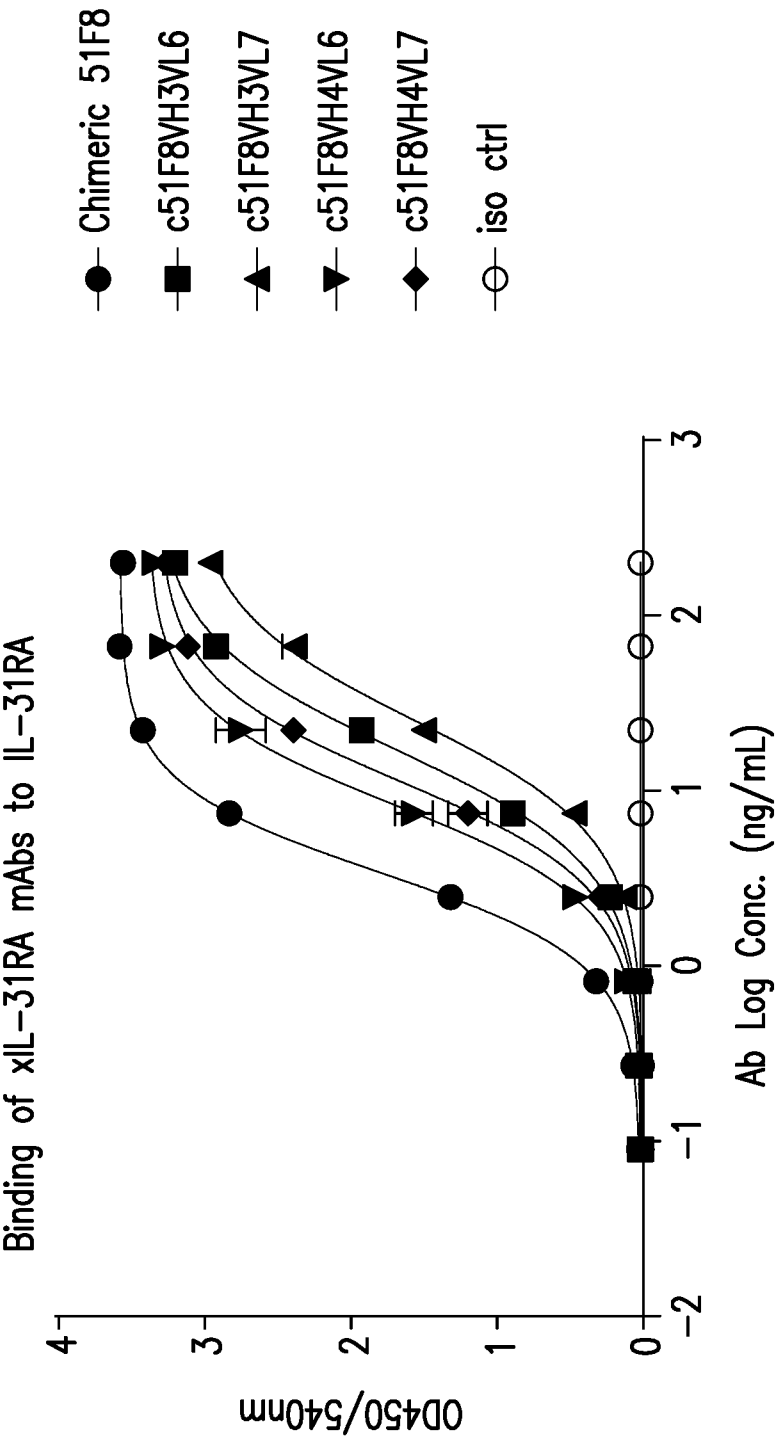
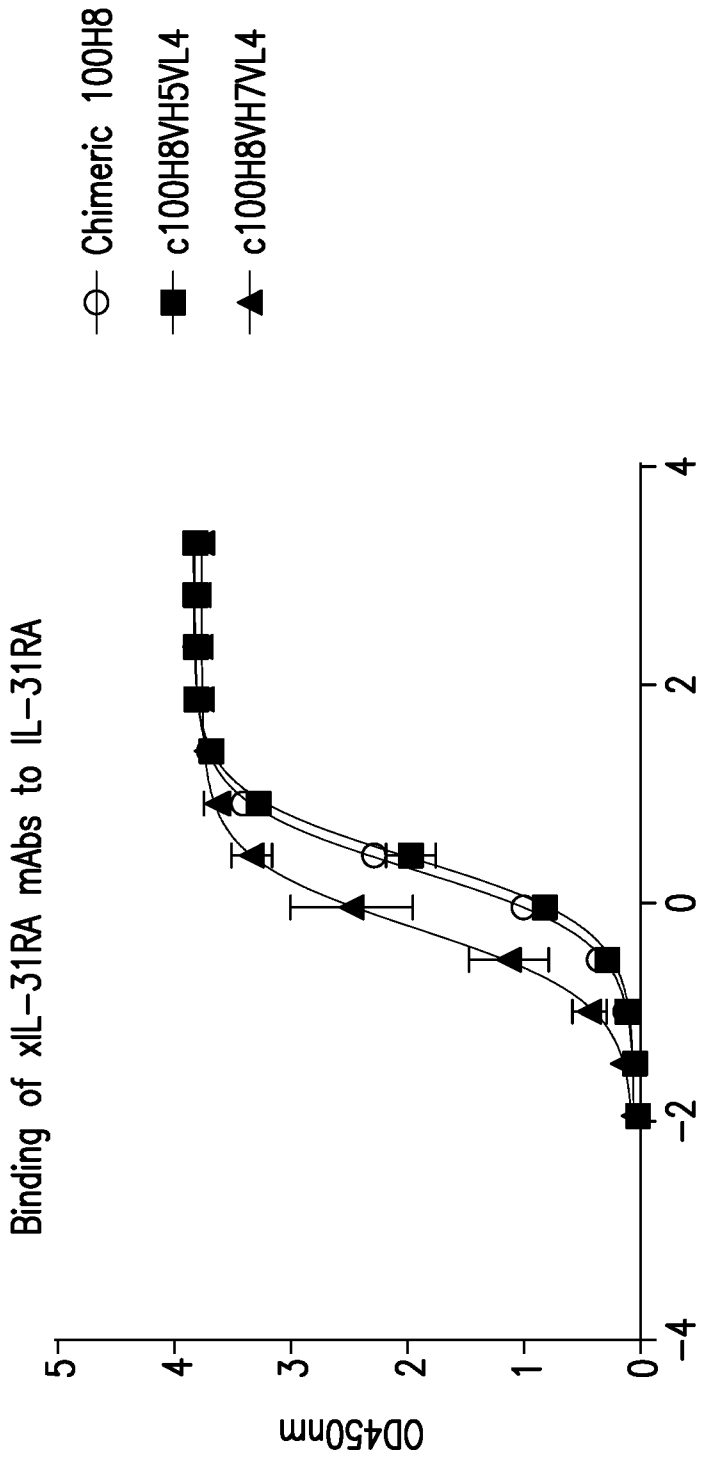

224G3 Epitope
478
487
SYSLQVMASTNAGGTNGTKIN
N
C
cIL-31RA
Cross-Link

FIG.6E



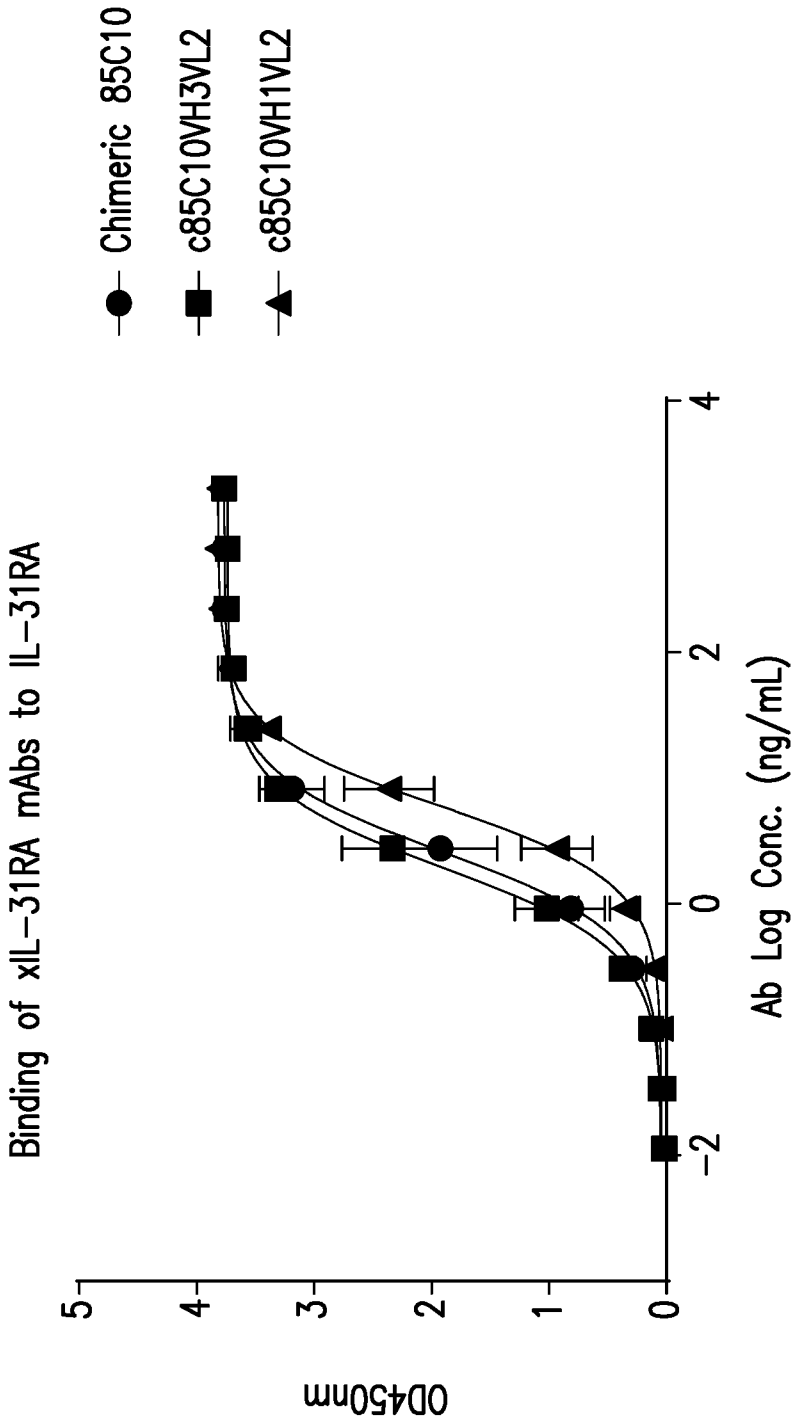
	Chimeric 51F8	c51F8VH3VL6	c51F8VH3VL7	c51F8VH4VL6	c51F8VH4VL7
EC50	3.417	16.94	25.02	8.306	11.33

FIG. 7A



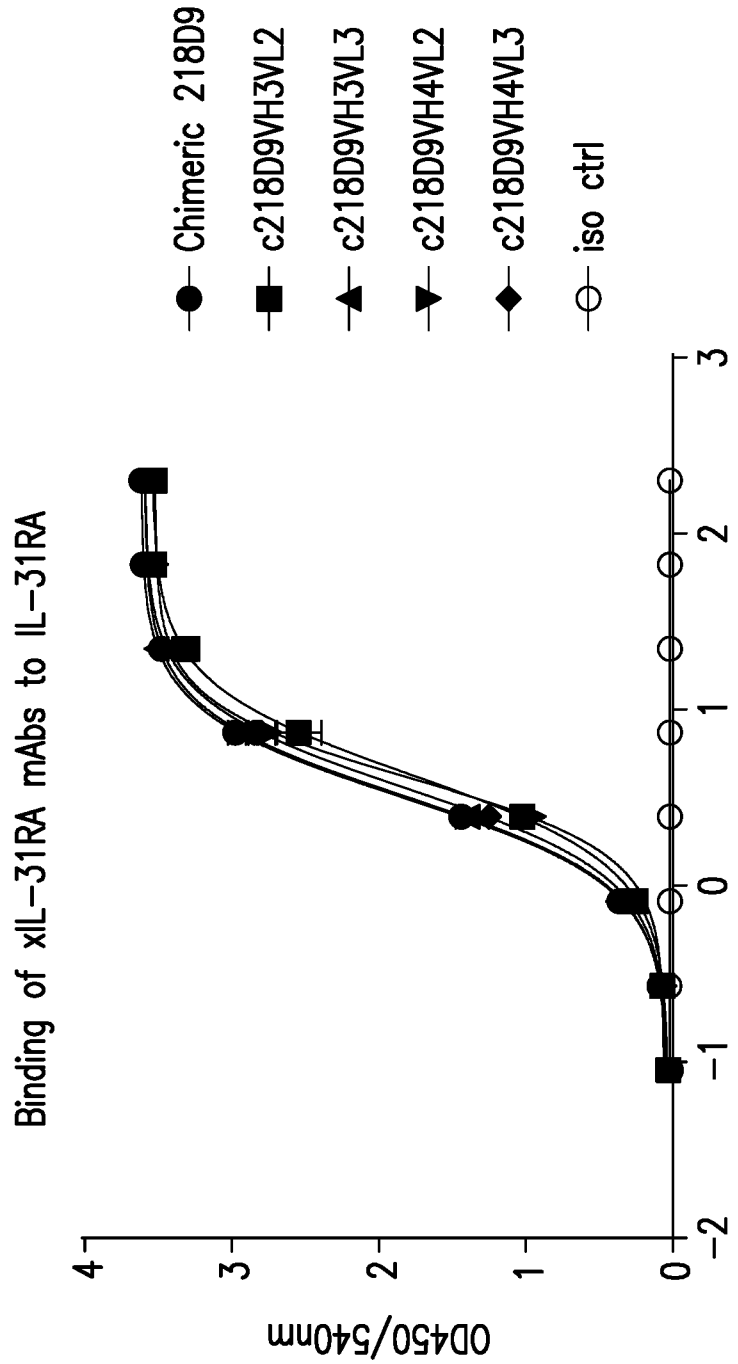
	Chimeric 100H8	c100H8VH5VL4	c100H8VH7VL4
EC50	2.021	2.561	0.5810

FIG.7B



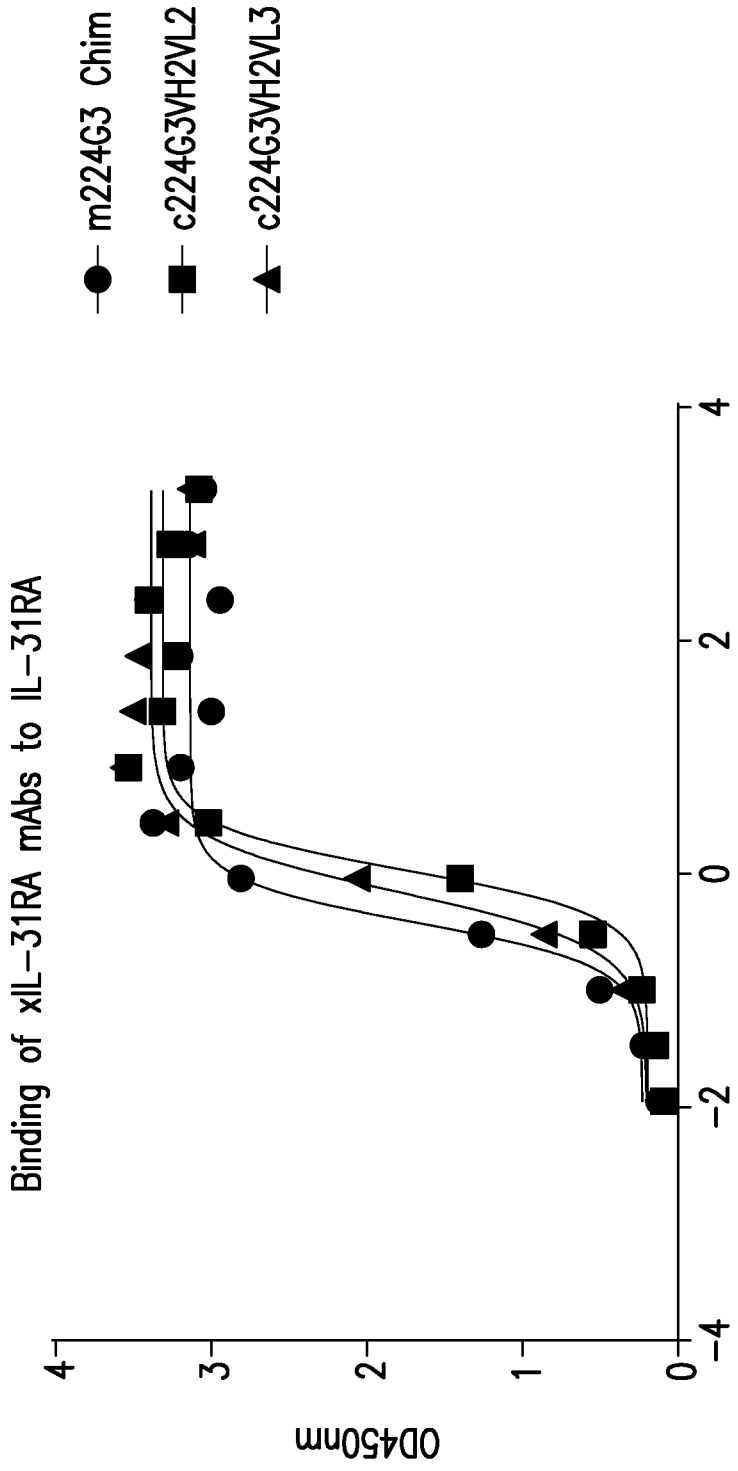
	Chimeric 85C10	c85C10VH3VL2	c85C10VH1VL2
EC50	2.583	1.910	6.002

FIG. 7C



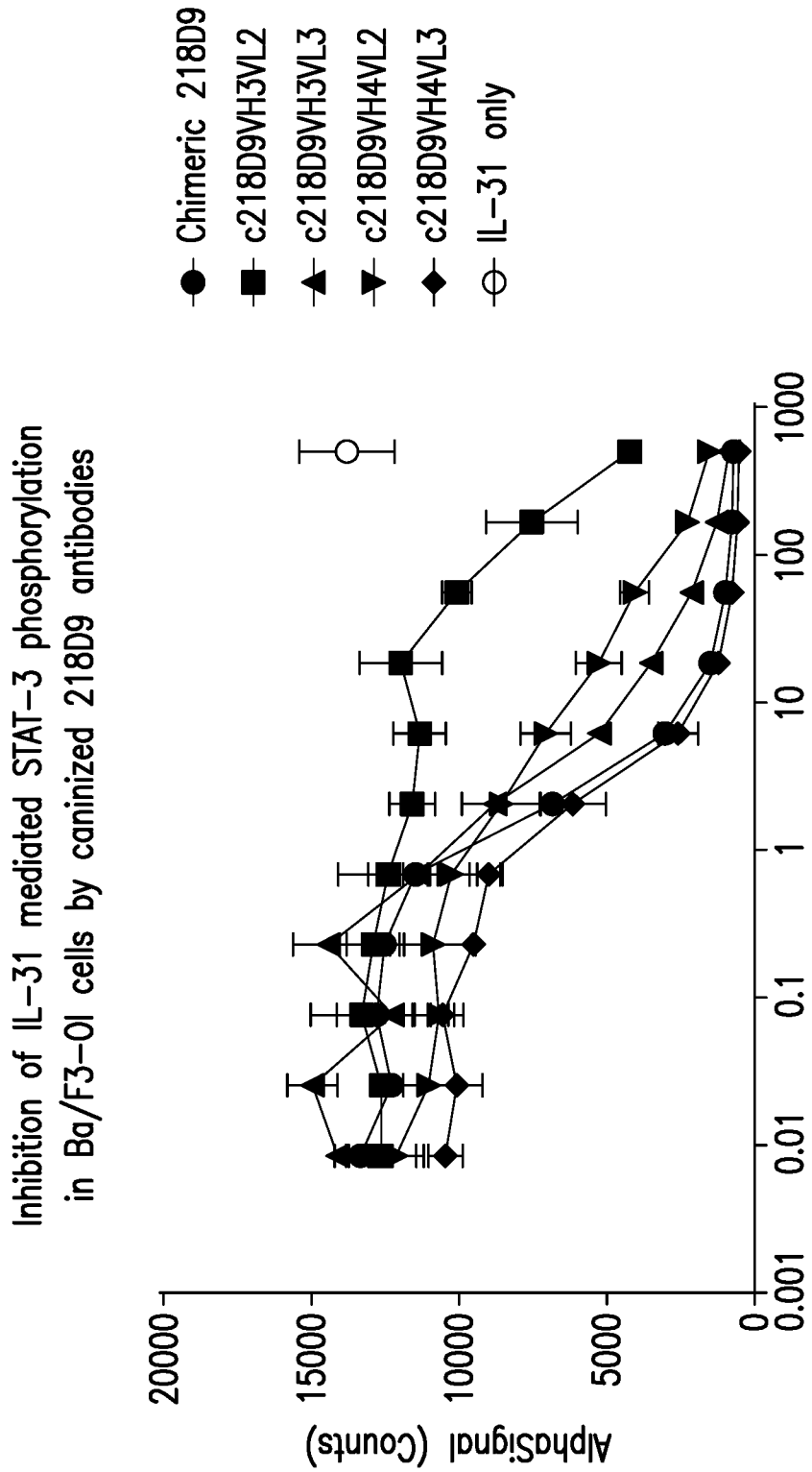
	Chimeric 218D9	c218D9VH3VL2	c218D9VH3VL3	c218D9VH4VL2	c218D9VH4VL3
EC50	3.153	4.289	3.216	4.096	3.580

FIG. 7D



	m224G3 Chim	c224G3VH2VL2	c224G3VH2VL3
EC50	0.3828	1.078	0.6985

FIG.7E



	Chimeric 218D9	c218D9VH3VL2	c218D9VH3VL3	c218D9VH4VL2	c218D9VH4VL3
IC50	2.237	226.2	2.972	8.265	2.498

FIG.8

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2022/086040

A. CLASSIFICATION OF SUBJECT MATTER
INV. C07K16/28 C07K14/715
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, BIOSIS, EMBASE, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 2020/116560 A1 (BICA THERAPEUTICS INC [JP]) 11 June 2020 (2020-06-11) the translation provided in EP3892632 is referred to -& EP 3 892 632 A1 (BICA THERAPEUTICS INC [JP]) 13 October 2021 (2021-10-13) e.g. paragraphs 11,12,39-41; examples 1 and 2; table 1; the whole document	1-29
A	WO 2020/142625 A2 (INVETX INC [US]) 9 July 2020 (2020-07-09) e.g. paragraphs 48,50,119; the whole document	1-29

-/--

[x] Further documents are listed in the continuation of Box C.

[x] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance;; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance;; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
15 March 2023	23/03/2023

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Gruber, Andreas

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Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2022/086040

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2021/123092 A1 (INTERVET INT BV [NL]; INTERVET INC [US]) 24 June 2021 (2021-06-24) e.g. claims 1,10,11; the whole document	1-29
A	WO 2018/073185 A1 (VETOQUINOL SA [FR]) 26 April 2018 (2018-04-26) e.g. claims 1, 42; the whole document	1-29
X	CN 110 563 844 A (UNIV HUAZHONG AGRICULTURAL) 13 December 2019 (2019-12-13) e.g. claims 1-4; the whole document	1-29
X	CA 3 161 496 A1 (INTERVET INT BV [NL]) 24 June 2021 (2021-06-24) e.g. claims 1,11; the whole document	1-29

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INTERNATIONAL SEARCH REPORT

International application No.

PCT/EP2022/086040

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:

   a. [X] forming part of the international application as filed.

   b. [ ] furnished subsequent to the international filing date for the purposes of international search (Rule 13*ter*.1(a)).

      [ ] accompanied by a statement to the effect that the sequence listing does not go beyond the disclosure in the international application as filed.

2. [ ] With regard to any nucleotide and/or amino acid sequence disclosed in the international application, this report has been established to the extent that a meaningful search could be carried out without a WIPO Standard ST.26 compliant sequence listing.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2022/086040

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2020116560 A1	11-06-2020	CN 113227134 A	06-08-2021
		EP 3892632 A1	13-10-2021
		JP WO2020116560 A1	21-10-2021
		US 2022048981 A1	17-02-2022
		WO 2020116560 A1	11-06-2020
EP 3892632 A1	13-10-2021	CN 113227134 A	06-08-2021
		EP 3892632 A1	13-10-2021
		JP WO2020116560 A1	21-10-2021
		US 2022048981 A1	17-02-2022
		WO 2020116560 A1	11-06-2020
WO 2020142625 A2	09-07-2020	AU 2020205073 A1	19-08-2021
		BR 112021012513 A2	14-09-2021
		CN 113544153 A	22-10-2021
		EP 3906259 A2	10-11-2021
		MA 54657 A	10-11-2021
		SG 11202106478U A	29-07-2021
		US 2020216536 A1	09-07-2020
		US 2020362035 A1	19-11-2020
		WO 2020142625 A2	09-07-2020
WO 2021123092 A1	24-06-2021	AU 2020406738 A1	23-06-2022
		AU 2020407269 A1	16-06-2022
		BR 112022011629 A2	13-09-2022
		BR 112022011669 A2	06-09-2022
		CA 3161496 A1	24-06-2021
		CA 3161728 A1	24-06-2021
		CN 114829396 A	29-07-2022
		CN 114829397 A	29-07-2022
		EP 4077379 A1	26-10-2022
		EP 4077380 A1	26-10-2022
		JP 2023506922 A	20-02-2023
		JP 2023506925 A	20-02-2023
		WO 2021123092 A1	24-06-2021
		WO 2021123094 A1	24-06-2021
WO 2018073185 A1	26-04-2018	CA 3040823 A1	26-04-2018
		CN 110114369 A	09-08-2019
		EP 3526246 A1	21-08-2019
		US 2020181258 A1	11-06-2020
		WO 2018073185 A1	26-04-2018
CN 110563844 A	13-12-2019	NONE	
CA 3161496 A1	24-06-2021	AU 2020406738 A1	23-06-2022
		AU 2020407269 A1	16-06-2022
		BR 112022011629 A2	13-09-2022
		BR 112022011669 A2	06-09-2022
		CA 3161496 A1	24-06-2021
		CA 3161728 A1	24-06-2021
		CN 114829396 A	29-07-2022
		CN 114829397 A	29-07-2022
		EP 4077379 A1	26-10-2022
		EP 4077380 A1	26-10-2022
		JP 2023506922 A	20-02-2023
		JP 2023506925 A	20-02-2023
		WO 2021123092 A1	24-06-2021

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2022/086040

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
		WO 2021123094 A1	24-06-2021