Claims

1. A method of producing a non-immunogenic immunoglobulin for administration to a target species, the method comprising the steps of:

- identifying a donor immunoglobulin from a species other than the target species, wherein the donor immunoglobulin has binding specificity to a target epitope present in the target species,
- determining an amino acid sequence of framework regions of heavy and/or light chain variable domains of the donor immunoglobulin,
- comparing each amino acid residue of the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin with an amino acid residue present at a corresponding position in an amino acid sequence of framework regions of one or more immunoglobulins derived from the target species to identify one or more amino acid residue within the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin that is not present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species, and
- substituting the one or more identified amino acid residues present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin, but not present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species, with an amino acid residue which is present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species.

2. The method as claimed in claim 1 wherein each amino acid residue of the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin is compared with a pool of amino acid residues comprising amino acid residues present at the corresponding position

in amino acid sequences of framework regions of a plurality of immunoglobulins derived from the target species.

3. The method as claimed in claim 1 or 2 wherein substitution of an amino acid residue present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin is undertaken using the principle of conservative substitution.

4. The method as claimed in claim 3 wherein conservative substitution comprises homologous substitution of the amino acid residue present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin with an amino acid residue which is present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species.

5. The method as claimed in claim 4 wherein the amino acid residue present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin is replaced with a conserved amino acid residue which has common characteristics with the amino acid which is being substituted, such as at least one of a similar side chain, a similar charge and similar hydrophobicity.

6. The method as claimed in claim 5 wherein the substituted amino acid residue and the replacement amino acid residue are both members of the same amino acid group, wherein the groups comprise:

- (1) non-polar residues: Ala (A), VaI (V), Leu (L), Ile (I), Pro (P), Phe (F), Trp (W), Met (M);
- (2) uncharged polar residues: GIy (G), Ser (S), Thr (T), Cys (C), Tyr (Y), Asn (N), Gln (Q); and
- (3) acidic residues: Asp (D), GIu (E); and (4) basic: Lys (K), Arg (R), His (H).

7. The method as claimed in claim 5 wherein the substituted amino acid residue and the replacement amino acid residue are both members of the same amino acid group, wherein the groups comprise:

- (1) aromatic: Phe (F), Trp (W), Tyr (Y);
- (2) apolar: Leu (L), Val (V), Ile (I), Ala (A), Met (M);
- (3) aliphatic: Ala (A), Val (V), Leu (L), Ile (I);
- (4) acidic: Asp (D), Glu (E);
- (5) basic: His (H), Lys (K), Arg (R); and
- (6) polar: Gln (Q), Asn (N), Ser (S), Thr (T), Tyr (Y).

8. The method as claimed in claim 5 wherein the substituted amino acid residue and the replacement amino acid residue are both members of the same amino acid group, wherein the groups comprise:

- (1) hydrophobic: Met (M), Ala (A), VaI (V), Leu (L), Ile (I);
- (2) neutral hydrophilic: Cys (C), Ser (S), Thr (T), Asn (N), Gin (Q);
- (3) acidic: Asp (D), GIu (E);
- (4) basic: His (H), Lys (K), Arg R);
- (5) residues that influence chain orientation: Gly (G), Pro (P); and
- (6) aromatic: Trp (W), Tyr (Y), Phe (F).

9. The method as claimed in claim 3 wherein conservative substitution comprises substitution of the amino acid residue present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin with a consensus amino acid residue which is present at the corresponding position in the amino acid sequence of the framework regions of the one or more immunoglobulins derived from the target species.

10. The method as claimed in any preceding claim further comprising the step of replacing at least one constant domain of the heavy and/or light chain of the donor immunoglobulin with a constant domain of a heavy and/or light chain derived from an immunoglobulin derived from the target species.

11. The method as claimed in claim 10 wherein the target species derived constant domains are of the antibody subtype Immunoglobulin G (IgG).

12. The method as claimed in any preceding claim wherein the target species is a mammalian target species.

13. The method as claimed in claim 12 wherein the mammalian species is a companion animal selected from the group consisting of a dog, a cat and a horse.

14. The method as claimed in claim 12 wherein the mammalian species is a human.

15. The method as claimed in any preceding claim wherein any amino acid residue which is present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin and also present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species is not substituted.

16. A composition containing an immunoglobulin produced according to any one of claims 1 to 15, or an antigen binding fragment thereof.

17. A pharmaceutical composition comprising an immunoglobulin produced according to any one of claims 1 to 15, or an antigen binding fragment thereof, and at least one pharmaceutically acceptable diluent or carrier.

18. Use of an immunoglobulin produced according to any one of claims 1 to 15, or an antigen binding fragment thereof, in the preparation of a medicament for the treatment of prevention of disease.

19. An immunoglobulin produced according to any one of claims 1 to 15, or an antigen binding fragment thereof, for use in the treatment or prevention of disease.

20. A method for treating disease in a subject in need thereof comprising the steps of administering a therapeutically effective amount of an immunoglobulin produced according to any one of claims 1 to 15, or an antigen binding fragment thereof, to the subject.

21. An immunoglobulin for administration to a target species, comprising:

- light and heavy chain constant domains derived from an immunoglobulin of the target species,
- complementary determining regions (CDRs) derived from a donor immunoglobulin wherein the donor immunoglobulin specifically binds to a ligand which is present in both the donor and target species, and
- framework regions derived from the donor immunoglobulin wherein any amino acid residues not also present at a corresponding position in any immunoglobulin from the target species are substituted with an amino acid which is found in the corresponding position in the target species.

22. The immunoglobulin as claimed in claim 21 wherein no amino acid substitutions are made to the CDR regions of the antibody.

23. The immunoglobulin as claimed in claim 21 or 22 wherein no more than 7 consecutive amino acid residues of the framework regions derived from the donor immunoglobulin are substituted with residues from the target species.

24. The immunoglobulin as claimed in claim 21 or 22 wherein no more than 5 consecutive amino acid residues of the framework regions derived from the donor immunoglobulin are substituted with residues from the target species.

25. The immunoglobulin as claimed in claim 21 or 22 wherein no more than 3 consecutive amino acid residues of the framework regions derived from the donor immunoglobulin are substituted with residues from the target species.

26. The immunoglobulin as claimed in any one of claims 21 to 23 wherein the target species is a mammalian species.

27. The immunoglobulin as claimed in claim 26 wherein the mammalian species is selected from the group consisting of a human, a canine, a feline and an equine.

28. A pharmaceutical composition comprising the immunoglobulin of any one of claims 21 to 27, or an antigen binding fragment thereof, and at least one pharmaceutically acceptable diluent or carrier.

29. Use of an immunoglobulin according to any one of claims 21 to 27, or an antigen binding fragment thereof, in the preparation of a medicament for the treatment of prevention of disease.

30. An immunoglobulin according to any one of claims 21 to 27, or an antigen binding fragment thereof, for use in the treatment or prevention of disease.

31. A method for treating disease in a subject in need thereof comprising the steps of administering a therapeutically effective amount of an immunoglobulin produced according to any one of claims 21 to 27, or an antigen binding fragment thereof, to the subject.

32. A neutralising antibody or an antigen binding fragment thereof which is capable of specifically binding to human nerve growth factor (NGF) wherein the antibody or antigen binding fragment comprises a light chain variable region comprising the amino acid sequence of SEQ ID NO:13 or an amino acid sequence which has an identity of at least 85% thereto and/or a heavy chain variable region

comprising the amino acid sequence of SEQ ID NO:14 or an amino acid sequence which has an identity of at least 85% thereto.

33. The antibody or antigen binding fragment thereof as claimed in claim 32 wherein the light chain comprises the amino acid sequence of SEQ ID NO:25, or an amino acid sequence which has an identity of at least 85% thereto.

34. The antibody or antigen binding fragment thereof as claimed in claim 32 or 33 wherein the heavy chain comprises the amino acid sequence of SEQ ID NO:24, or an amino acid sequence which has a sequence identity of at least 85% thereto.

35. A pharmaceutical composition comprising the antibody or antigen binding fragment thereof as claimed in any one of claims 32 to 34 and at least one pharmaceutically acceptable diluent or carrier.

36. An isolated nucleic acid that encodes an antibody or antigen binding fragment according to any one of claims 32 to 34.

37. An isolated nucleic acid which encodes a light chain variable region of a neutralising antibody which is capable of specifically binding to human nerve growth factor (NGF), wherein the light chain variable region comprises the amino acid sequence of SEQ ID NO:13 or an amino acid sequence which has an identity of at least 85% thereto.

38. An isolated nucleic acid which encodes a light chain of a neutralising antibody which is capable of specifically binding to human nerve growth factor (NGF), wherein the light chain comprises the amino acid sequence of SEQ ID NO:25 or an amino acid sequence which has an identity of at least 85% thereto.

39. An isolated nucleic acid which encodes a heavy chain variable region of a neutralising antibody which is capable of specifically binding to human nerve

growth factor (NGF), wherein the heavy chain variable region comprises the amino acid sequence of SEQ ID NO:14 or an amino acid sequence which has an identity of at least 85% thereto.

40. An isolated nucleic acid which encodes a heavy chain of a neutralising antibody which is capable of specifically binding to human nerve growth factor (NGF), wherein the heavy chain comprises the amino acid sequence of SEQ ID NO:24 or an amino acid sequence which has an identity of at least 85% thereto.

41. The isolated nucleic acid as claimed in any one of claims 36 to 40 which is operably linked to one or more regulatory sequences.

42. An expression vector comprising a nucleic acid as claimed in any one of claims 36 to 40.

43. The expression vector as claimed in claim 42 further comprising one or more regulatory sequences.

44. The expression vector as claimed in claim 42 or 43 wherein the expression vector is a plasmid or a retroviral vector.

45. A host cell incorporating the expression vector as claimed in any one of claims 42 to 44.

46. A method for producing a humanised NGF neutralising antibody or antigen binding fragment thereof, the method comprising the step of culturing a host cell as claimed in claim 45 to allow the cell to express a humanised anti-NGF neutralising antibody or antigen binding fragment thereof.

47. A method for treating, ameliorating or inhibiting pain in a human in need thereof, the method comprising the step of administering to the human a

therapeutically effective amount of an antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one of claims 42 to 44.

48. An antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one of claims 42 to 44 for use in the treatment or prevention of pain in a human.

49. The antibody, antigen binding fragment, pharmaceutical composition, nucleic acid or expression vector as claimed in claim 48 wherein the pain is selected from the group consisting of acute pain, chronic pain, peri-operative pain, post-operative pain, inflammatory pain, pruritic pain, pain resulting from orthopaedic surgery, pain associated with soft tissue surgery, chronic pain associated with cancer or a cancerous condition and pain associated with, or resulting from, rheumatoid arthritis, immune mediated polyarthritis or osteoarthritis.

50. An antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one of claims 42 to 44 for use in the treatment of arthritis in a human.

51. The antibody, antigen binding fragment, pharmaceutical composition, nucleic acid or expression vector as claimed in claim 50 wherein the arthritis is selected from the group consisting of osteoarthritis, immune mediated polyarthritis and rheumatoid arthritis.

52. Use of antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one

of claims 42 to 44 in the preparation of a medicament for the treatment or prevention of pain in a human.

53. Use of antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one of claims 42 to 44 in the preparation of a medicament for the treatment or prevention of arthritis in a human.

54. A method for treating or preventing arthritis in a human, the method comprising the step of administering to the human a therapeutically effective amount of an antibody or antigen binding fragment according to any one of claims 32 to 34, a pharmaceutical composition as claimed in claim 35, a nucleic acid as claimed in any one of claims 36 to 41 or an expression vector as claimed in any one of claims 42 to 44.

55. A cell line or a derivative or progeny cell thereof that produces anti-human NGF neutralising monoclonal antibodies or fragments thereof according to any one of claims 32 to 34.

56. A neutralising antibody or an antigen binding fragment thereof which is capable of specifically binding to canine nerve growth factor (NGF) wherein the antibody or antigen binding fragment comprises a light chain variable region comprising the amino acid sequence of SEQ ID NO:1 or an amino acid sequence which has an identity of at least 85% thereto and/or a heavy chain variable region comprising the amino acid sequence of SEQ ID NO:69 or an amino acid sequence which has an identity of at least 85% thereto.

57. The antibody or antigen binding fragment thereof as claimed in claim 56 wherein the light chain comprises the amino acid sequence of SEQ ID NO:7, or an amino acid sequence which has an identity of at least 85% thereto.

58. The antibody or antigen binding fragment thereof as claimed in claim 56 or 57 wherein the heavy chain comprises the amino acid sequence of SEQ ID NO:70, or an amino acid sequence which has a sequence identity of at least 85% thereto.

59. A pharmaceutical composition comprising the antibody or antigen binding fragment thereof as claimed in any one of claims 56 to 58 and at least one pharmaceutically acceptable diluent or carrier.

60. An isolated nucleic acid that encodes an antibody or antigen binding fragment according to any one of claims 56 to 58.

61. An isolated nucleic acid which encodes a heavy chain variable region of a neutralising antibody which is capable of specifically binding to canine nerve growth factor (NGF), wherein the heavy chain variable region comprises the amino acid sequence of SEQ ID NO:69 or an amino acid sequence which has an identity of at least 85% thereto.

62. An isolated nucleic acid which encodes a heavy chain of a neutralising antibody which is capable of specifically binding to canine nerve growth factor (NGF), wherein the heavy chain comprises the amino acid sequence of SEQ ID NO:70 or an amino acid sequence which has an identity of at least 85% thereto.

63. The isolated nucleic acid as claimed in any one of claims 60 to 62 which is operably linked to one or more regulatory sequences.

64. An expression vector comprising a nucleic acid as claimed in any one of claims 60 to 62.

65. The expression vector as claimed in claim 64 further comprising one or more regulatory sequences.

66. The expression vector as claimed in claim 64 or 65 wherein the expression vector is a plasmid or a retroviral vector.

67. A host cell incorporating the expression vector as claimed in any one of claims 64 to 66.

68. A method for producing a caninised NGF neutralising antibody or antigen binding fragment thereof, the method comprising the step of culturing a host cell as claimed in claim 67 to allow the cell to express a caninised anti-NGF neutralising antibody or antigen binding fragment thereof.

69. A method for treating, ameliorating or inhibiting pain in a canine in need thereof, the method comprising the step of administering to the canine a therapeutically effective amount of an antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66.

70. An antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66 for use in the treatment or prevention of pain in a canine.

71. The antibody, antigen binding fragment, pharmaceutical composition, nucleic acid or expression vector as claimed in claim 70 wherein the pain is selected from the group consisting of acute pain, chronic pain, peri-operative pain, post-operative pain, inflammatory pain, pruritic pain, pain resulting from orthopaedic surgery, pain associated with soft tissue surgery, chronic pain associated with cancer or a cancerous condition and pain associated with, or resulting from, rheumatoid arthritis, immune mediated polyarthritis or osteoarthritis.

72. An antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66 for use in the treatment of arthritis in a canine.

73. The antibody, antigen binding fragment, pharmaceutical composition, nucleic acid or expression vector as claimed in claim 72 wherein the arthritis is selected from the group consisting of osteoarthritis, immune mediated polyarthritis and rheumatoid arthritis.

74. Use of antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66 in the preparation of a medicament for the treatment or prevention of pain in a canine.

75. Use of antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66 in the preparation of a medicament for the treatment or prevention of arthritis in a canine.

76. A method for treating or preventing arthritis in a canine, the method comprising the step of administering to the canine a therapeutically effective amount of an antibody or antigen binding fragment according to any one of claims 56 to 58, a pharmaceutical composition as claimed in claim 59, a nucleic acid as claimed in any one of claims 60 to 63 or an expression vector as claimed in any one of claims 64 to 66.

77. A cell line or a derivative or progeny cell thereof that produces anti-canine NGF neutralising monoclonal antibodies or fragments thereof according to any one of claims 56 to 58.

Claims

1. A method of modifying a donor immunoglobulin for use in a target species, the method comprising the steps of:

- identifying a donor immunoglobulin from a species other than the target species, wherein the donor immunoglobulin has binding specificity to a target epitope present in the target species,
- determining an amino acid sequence of framework regions of heavy and/or light chain variable domains of the donor immunoglobulin,
- comparing each amino acid residue of the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin with an amino acid residue present at a corresponding position in an amino acid sequence of framework regions of one or more immunoglobulins derived from the target species to identify one or more amino acid residue within the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin that is not present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species, and
- substituting the one or more identified amino acid residues present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin, but not present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species, with an amino acid residue which is present at the corresponding position in the amino acid sequence of the framework regions of at least one of the one or more immunoglobulins derived from the target species;
- wherein the modified donor immunoglobulin does not contain any amino acid in any position within the framework regions of the heavy and/or light chain variable domains which would be foreign at that position in one or more immunoglobulins derived from the target species.

2. The method as claimed in claim 1, wherein substitution of an amino acid residue present in the amino acid sequence of the framework regions of the heavy and/or light chain variable domains of the donor immunoglobulin is undertaken using the principle of conservative substitution.

3. The method as claimed in claim 1 or claim 2, further comprising the step of replacing at least one constant domain of the heavy and/or light chain of the donor immunoglobulin with a constant domain of a heavy and/or light chain derived from an immunoglobulin derived from the target species.

4. The method as claimed in any preceding claim wherein the target species is a mammalian target species.

5. The method as claimed in claim 4 wherein the mammalian species is a companion animal selected from the group consisting of a dog, a cat and a horse.

6. The method as claimed in claim 4 wherein the mammalian species is a human.

7. A neutralising antibody or an antigen binding fragment thereof which is capable of specifically binding to human nerve growth factor (NGF) wherein the antibody or antigen binding fragment comprises a light chain variable region comprising the amino acid sequence of SEQ ID NO:13 or an amino acid sequence which has an identity of at least 85% thereto and/or a heavy chain variable region comprising the amino acid sequence of SEQ ID NO:14 or an amino acid sequence which has an identity of at least 85% thereto, wherein the antibody or an antigen binding fragment thereof does not contain any amino acid in any position within the framework regions of the heavy and/or light chain variable domains which would be foreign at that position in one or more immunoglobulins derived from a human.

8. The antibody or antigen binding fragment thereof as claimed in claim 7 wherein the light chain comprises the amino acid sequence of SEQ ID NO:25, or an amino acid sequence which has an identity of at least 85% thereto.

9. The antibody or antigen binding fragment thereof as claimed in claim 7 or claim 8 wherein the heavy chain comprises the amino acid sequence of SEQ ID NO:24, or an amino acid sequence which has a sequence identity of at least 85% thereto.

10. A pharmaceutical composition comprising the antibody or antigen binding fragment thereof as claimed in any one of claims 7 to 9 and at least one pharmaceutically acceptable diluent or carrier.

11. The antibody or antigen binding fragment as claimed in any one of claims 7 to 9, or the pharmaceutical composition as claimed in claim 10, for use in the treatment or prevention of pain in a human.

12. The antibody or antigen binding fragment as claimed in any one of claims 7 to 9, or the pharmaceutical composition as claimed in claim 10, for use in the treatment of arthritis in a human.

13. Use of the antibody or antigen binding fragment as claimed in any one of claims 7 to 9, in the preparation of a medicament for the treatment or prevention of pain in a human.

14. Use of the antibody or antigen binding fragment as claimed in any one of claims 7 to 9 in the preparation of a medicament for the treatment or prevention of arthritis in a human.