Claims

1. A caninised antibody or an antigen binding fragment thereof which binds specifically to canine tumour necrosis factor .

2. The caninised antibody or antigen binding fragment thereof as claimed in claim 1 wherein the antibody is prepared by a method comprising the steps of:

- providing a donor antibody from a species other than a canine, wherein the donor antibody has binding specificity for tumour necrosis factor;

- comparing each amino acid residue of the amino acid sequence of framework regions of the donor antibody with each amino acid residue present at a corresponding position in the amino acid sequence of framework regions of one or more canine antibodies to identify one or more amino acid residues within the amino acid sequence of the framework regions of the donor antibody that differ from one or more amino acid residues at the corresponding position within the amino acid sequence of framework regions of the one or more canine antibodies; and

- substituting the one or more identified amino acid residues in the donor antibody with the one or more amino acid residues present at the corresponding position in the one or more canine antibodies.

3. The antibody or antigen binding fragment thereof as claimed in claim 2 wherein the step of substituting the one or more identified amino acid residues comprises substituting the one or more identified amino acid residues with the one or more amino acid residues present at the corresponding position which have the highest homology to the one or more substituted amino acid residues.

4. The antibody or antigen binding fragment thereof as claimed in claim 2 or 3 wherein the method further comprises the step of replacing constant domains of the heavy chain and/or light chain of the donor antibody with constant domains of a heavy and/or light chain from a canine antibody.

5. The antibody or antigen binding fragment thereof as claimed in any one of claims 1 to 4 wherein the antibody or antigen binding fragment thereof 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:2 or an amino acid sequence which has an identity of at least 85% thereto.

6. The antibody or antigen binding fragment as claimed in claim 5 wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO:3 or an amino acid sequence which has an identity of at least 85% thereto and/or a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6 and SEQ ID NO:7, or an amino acid sequence which has a sequence identity of at least 85% thereto.

7. The antibody or antigen binding fragment as claimed in any one of claims 1 to 5 wherein the antibody comprises a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:4, aglycosylated SEQ ID NO:4, aglycosylated SEQ ID NO:5, aglycosylated SEQ ID NO:6, aglycosylated SEQ ID NO:7 and SEQ ID NO:7, or an amino acid sequence which has a sequence identity of at least 85% thereto.

8. The antibody or antigen binding fragment as claimed in any one of claims 1 to 5 wherein the antibody comprises a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:5 and SEQ ID NO:6, or an amino acid sequence which has a sequence identity of at least 85% thereto.

9. The antibody or antigen binding fragment thereof as claimed in any one of claims 1 to 4 wherein the antibody or antigen binding fragment comprises a light chain variable region comprising at least one of:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:10,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:11,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:12, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:13,

and/or a heavy chain variable region comprising at least one of:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:14,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:15,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:16, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:17.

10. The antibody or antigen binding fragment thereof as claimed in claim 9 which comprises a light chain variable domain having an FR1 region of SEQ ID NO:10 which has been modified by one or more of the amino acid substitutions selected from the group consisting of amino acid residue T at position 5 (T5) is replaced by the amino acid residue M or I, S7 is T, A9 is L or P, S12 is A, L13 is V, S14 is T or R, Q15 is P or R, E16 is D, K18 is E, A, P, T or L, V19 is A, T20 is S, T22 is S or Y and C23 is Y.

11. The antibody or antigen binding fragment thereof as claimed in claim 9 or 10 which comprises a light chain variable domain having the FR2 region of SEQ ID NO:11 which has been modified by one or more of the amino acid substitutions selected from the group consisting of Y2 is F, I or L, Q3 is R, L or I, Q4 is H, K5 is R, P6

is S or A, G7 is D, A9 is S, T or P, K11 is Q, E or R, L12 is R, P, G, A or S, I14 is L and Y15 is F, N, S, E or V.

12. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 11 which comprises a light chain variable domain having the FR3 region of SEQ ID NO:12 which has been modified by one or more of the amino acid substitutions selected from the group consisting of G1 is A, V2 is A, P3 is S, S4 is D, F6 is L or V, S7 is I, G8 is A, T13 is A, D14 is E, T16 is S or R, L17 is F, T18 is R or K, S21 is R, G or T, L22 is V, P24 is A, E25 is D, G, I or N, V27 is A, T, G or S, A28 is G and V29 is I or L.

13. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 12 which comprises a light chain variable domain having the FR4 region of SEQ ID NO:13 has been modified by one or more of the amino acid substitutions selected from the group consisting of G2 is S, Q3 is A, P or T, G4 is E, T5 is P, K6 is Q or S, V7 is L or W, E8 is D or R and I9 is L.

14. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 13 which comprises a heavy chain variable domain having the FR1 region of SEQ ID NO:14 which has been modified by one or more of the amino acid substitutions selected from the group consisting of E1 is D or G, V2 is G, L, E, I or M, Q3 is H, R, A, V, E, K, L, P or S, L4 is V or P, V5 is A, L, E or M, E6 is Q or A, S7 is F, L or T, G9 is E, G10 is D, A, N, E or T, L11 is Q, R, V or W, V12 is A, I or M, Q13 is K, R or N, P14 is F or T, G15 is A, E or T, G16 is E or A, S17 is T or P, L18 is R, R19 is K, T, G or V, L20 is I or V, S21 is Y, A23 is V, L, I or E, A24 is T, V, G, I or S, S25 is P, G or T, G26 is D, R or T and F27 is L, I, S, D, T or V.

15. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 14 which comprises a heavy chain variable domain having the FR2 region of SEQ ID NO:15 which has been modified by one or more of the amino acid substitutions selected from the group consisting of W1 is C, V2 is I, A, F or L, Q4 is L

or H, A5 is S, T, G, P, V or D, P6 is L, G7 is E, R or L, K8 is R, E, G, A, M or Q, G9 is E, R, D, T or V, L10 is T, P, F or M, E11 is Q, H, D, L, P or R, W12 is L, C, S, Y, F or M, V13 is L, I or F and S14 is A, T, G or L.

16. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 15 which comprises a heavy chain variable domain having the FR3 region of SEQ ID NO:16 which has been modified by one or more of the amino acid substitutions selected from the group consisting of R1 is Q, F2 is V or L, T3 is A, I or S, I4 is V, L, M or T, S5 is A, F or T, R6 is K, D7 is E or N, N8 is D, T, S, I or G, A9 is G, V, S, D, P or T, K10 is R, E, N, Q, G or M, N11 is D, S, K, H or R, S12 is T, M, I or A, L13 is V, M, A or I, Y14 is F, H, S or T, L15 is I, Q16 is H, E, D, R or A, M17 is L, N18 is D, S, T, H, K, P or R, S19 is G, D, R, N or T, L20 is V, R21 is T, G, K, S or I, A22 is V, D, T, S, G or P, E23 is D, A or V, T25 is A, S or M, A26 is G or V, V27 is M, I, L, F, T, K or Q, Y28 is H, Y29 is F or H, A31 is V, T, G, M, R, S, C or L and K32 is R, S, N, G, A, T, P, D, Q, V, E, I or M.

17. The antibody or antigen binding fragment thereof as claimed in any one of claims 9 to 16 which comprises a heavy chain variable domain having the FR4 region of SEQ ID NO:17 which has been modified by one or more of the amino acid substitutions selected from the group consisting of W1 is L, G2 is A or S, Q3 is P, H, R or D, T5 is A, S, I or N, L6 is S, Q, P or R, V7 is L, I or P, T8 is F, I, A, S, L, P or Y, V9 is A, S10 is A, C, P or T and S11 is L, A or P.

18. A chimeric antibody or an antigen binding fragment thereof which specifically binds to canine tumour necrosis factor wherein the antibody or antigen binding fragment comprises a light chain comprising the amino acid sequence of SEQ ID NO:8 or an amino acid sequence which has an identity of at least 85% thereto.

19. The antibody or antigen binding fragment as claimed in claim 18 wherein the antibody or antigen binding fragment comprises a heavy chain comprising the

amino acid sequence of SEQ ID NO:9 or an amino acid sequence which has an identity of at least 85% thereto.

20. The antibody or antigen binding fragment as claimed in claim 18 wherein the antibody or antigen binding fragment comprises a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:20, SEQ ID NO:21 and SEQ ID NO:22, or an amino acid sequence which has an identity of at least 85% thereto.

21. The antibody of antigen binding fragment as claimed in any one of claims 1 to 20 wherein the antibody or antigen binding fragment binds to canine TNF with a binding affinity (K_D) of $1x10^{-8}$ or less.

22. The antibody or antigen binding fragment thereof as claimed in any one of claims 1 to 21 wherein the binding of the antibody or antigen binding fragment to canine TNF inhibits the ability of canine TNF to bind to TNFR1 receptor.

23. The antigen binding fragment thereof as claimed in any one of claims 1 to 22 wherein the antigen binding fragment is selected from the group consisting of a single chain Fv (scFv) antibody fragment, a Fab antibody fragment, a Fab' antibody fragment and a $F(ab')_2$ antibody fragment.

24. The antibody or antigen binding fragment thereof as claimed in any one of claims 1 to 22 wherein the antibody is a multivalent antibody.

25. The antibody or antigen binding fragment thereof as claimed in any one of claims 1 to 22 wherein the antibody is a multispecific antibody.

26. A method for treating, ameliorating or preventing an immune mediated condition, the method comprising the steps of:

- providing a therapeutically effective amount of an antibody or antigen binding fragment as claimed in any one of claims 1 to 25, and
- administering the same to a canine in need thereof.

27. The method as claimed in claim 26 wherein the immune mediated condition is a chronic inflammatory disease.

28. The method as claimed in claim 27 wherein the chronic inflammatory disease is selected from the group consisting of immune mediated polyarthritis, rheumatoid arthritis, osteoarthritis, ankylosing spondylitis, Crohn's disease, ulcerative colitis, psoriasis and psoriatic arthritis, systemic vasculitis, atopic dermatitis, congestive heart failure, refractory uveitis and bronchial asthma.

29. The method as claimed in claim 26 wherein the immune mediated condition is a TNF-alpha related disorder selected from the group consisting of sepsis, septic shock, diabetes mellitus, Alzheimer's disease, Parkinson's disease, stroke, ischaemic heart disease and amyotrophic lateral sclerosis.

30. The method as claimed in any one of claims 26 to 29 further comprising the step of co-administering at least one further therapeutic agent.

31. The method as claimed in claim 30 wherein the at least one therapeutic agent is selected from the group consisting of methotrexate, a canine TNF fusion protein, soluble p55 or p75 TNF receptor or derivatives thereof, a chimeric or canine antibody to canine TNF, an anti-canine TNF antibody fragment, an analgesic, an NSAID, an opioid, a corticosteroid, a steroid and a nerve growth factor antagonist.

32. The method as claimed in claim 30 wherein the at least one therapeutic agent is selected from the group consisting of antibiotic, antifungal, antiprotozoal and antiviral therapeutic agents.

33. The method as claimed in claim 30 wherein the at least one therapeutic agent is selected from the group consisting of an inhibitor of a mediator of inflammation, a PGE-receptor antagonist, an immunosuppressive agent, cyclosporine, an anti-inflammatory glucocorticoid, an agent for use in the treatment of cognitive dysfunction or cognitive impairment, an anti-hypertensive, a compound used for the treatment of cardiovascular dysfunction, a diuretic, a vasodilator, a beta-adrenergic receptor antagonist, an angiotensin-II converting enzyme inhibitor, a calcium channel blocker and a HMG-CoA reductase inhibitor.

34. The method as claimed in any one of claims 26 to 33 wherein the antibody is administered to the canine at a dose ranging from about 0.01 mg/kg of body weight to about 10 mg/kg of body weight.

35. A pharmaceutical composition for treating, ameliorating or preventing an immune mediated condition comprising a therapeutically effective amount of an antibody or an antigen binding fragment thereof according to any one of claims 1 to 25 along with at least one pharmaceutically acceptable carrier, excipient or diluent.

36. The pharmaceutical composition as claimed in claim 35 further comprising at least at least one further therapeutic agent selected from the group consisting of methotrexate, a canine TNF fusion protein, soluble p55 or p75 TNF receptor or derivatives thereof, a chimeric or canine antibody to canine TNF, an anti-canine TNF antibody fragment, an analgesic, an NSAID, an opioid, a corticosteroid, a steroid and a nerve growth factor antagonist.

37. An isolated nucleic acid that encodes an antibody or antigen binding fragment according to any one of claims 1 to 25.

38. An isolated nucleic acid which encodes the light chain variable domain of an anti-canine TNF caninised antibody or antibody fragment having the amino acid

sequence of SEQ ID NO:1, or a light chain having the amino acid sequence of SEQ ID NO:3.

39. An isolated nucleic acid which encodes the heavy chain variable domain of an anti-canine TNF caninised antibody or antibody fragment having the amino acid sequence of SEQ ID NO:2 or a heavy chain having the amino acid sequence selected from the group consisting of SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6 and SEQ ID NO:7.

40. The isolated nucleic acid as claimed in any one of claims 37 to 39 which is operably linked to one or more regulatory sequences.

41. An expression vector comprising a nucleic acid as claimed in any one of claims 37 to 39.

42. The expression vector as claimed in claim 41 further comprising one or more regulatory sequences.

43. The expression vector as claimed in claim 41 or 42 wherein the expression vector is a plasmid or a retroviral vector.

44. A host cell incorporating the expression vector as claimed in any one of claims 41 to 43.

45. A method for producing an anti-canine TNF neutralising antibody, the method comprising the step of culturing a host cell as claimed in claim 44 to allow the cell to express a caninised anti-canine TNF neutralising antibody.

46. A method for treating, ameliorating or inhibiting an immune mediated condition in a canine, the method comprising the step of administering to the canine

a therapeutically effective amount of a nucleic acid according to any one of claims 37 to 39.

47. An antibody or antigen binding fragment according to any one of claims 1 to 25, a pharmaceutical composition according to claim 35 or 36, a nucleic acid as claimed in any one of claims 37 to 39 or an expression vector as claimed in any one of claims 41 to 43 for use in the treatment or prevention of a chronic inflammatory disease in a canine.

48. The antibody or antigen binding fragment as claimed in claim 47 wherein the chronic inflammatory disease is selected from the group consisting of rheumatoid arthritis, osteoarthritis and other polyarthritidies, immune mediated polyarthritis, ankylosing spondylitis, Crohn's disease and ulcerative colitis, psoriasis and psoriatic arthritis, systemic vasculitis, atopic dermatitis, congestive heart failure, refractory uveitis, bronchial asthma and allergic conditions.

49. An antibody or antigen binding fragment according to any one of claims 1 to 25, a pharmaceutical composition according to claim 35 or 36, a nucleic acid as claimed in any one of claims 37 to 39 or an expression vector as claimed in any one of claims 41 to 43 for use in the treatment of arthritis.

50. The antibody or antigen binding fragment as claimed in claim 49 wherein the arthritis is selected from the group consisting of osteoarthritis, immune mediated polyarthritis and rheumatoid arthritis.

51. Use of an antibody or antigen binding fragment according to any one of claims 1 to 25, a pharmaceutical composition according to claim 35 or 36, a nucleic acid as claimed in any one of claims 37 to 39 or an expression vector as claimed in any one of claims 41 to 43 in the preparation of a medicament for the treatment, amelioration or prevention of a chronic inflammatory disease in a canine.

52. Use of the antibody or antigen binding fragment as claimed in claim 51 wherein the chronic inflammatory disease is selected from the group consisting of rheumatoid arthritis, immune mediated polyuarthritis, osteoarthritis and other polyarthritidies, ankylosing spondylitis, Crohn's disease and ulcerative colitis, psoriasis, psoriatic arthritis, systemic vasculitis, atopic dermatitis, congestive heart failure, refractory uveitis, bronchial asthma and allergic conditions.

53. Use of an antibody or antibody binding fragment according to any one of claims 1 to 25, a pharmaceutical composition according to claim 35 or 36, a nucleic acid as claimed in any one of claims 37 to 39 or an expression vector as claimed in any one of claims 41 to 43 in the preparation of a medicament for the treatment, inhibition amelioration or prevention of arthritis in a canine.

54. Use of the antibody or antibody binding fragment as claimed in claim 53 wherein the arthritis is selected from the group consisting of osteoarthritis, immune mediated polyarthritis and rheumatoid arthritis.

55. A cell line or a derivative or progeny cell thereof that produces anti-canine TNF neutralising monoclonal antibodies, or fragments thereof according to any one of claims 1 to 25.

56. A kit for the treatment of a canine chronic inflammatory disease comprising an anti-canine TNF antibody according to any one of claims 1 to 25 and instructions for use of the same.

Claims

1. A method of preparing a caninised antibody or an antigen binding fragment thereof which binds specifically to canine tumour necrosis factor alpha, the method comprising the steps of:

- providing a donor antibody from a species other than a canine, wherein the donor antibody has binding specificity for tumour necrosis factor alpha;

- comparing each amino acid residue of the amino acid sequence of framework regions of the donor antibody with each amino acid residue present at a corresponding position in the amino acid sequence of framework regions of one or more canine antibodies to identify one or more amino acid residues within the amino acid sequence of the framework regions of the donor antibody that differ from one or more amino acid residues at the corresponding position within the amino acid sequence of framework regions of the one or more canine antibodies; and

- substituting the one or more identified amino acid residues in the donor antibody with the one or more amino acid residues present at the corresponding position in the one or more canine antibodies,

wherein the caninised antibody or antigen binding fragment thereof does not contain any amino acid in any position within the framework regions which would be foreign at that position in canines.

2. An antibody or an antigen binding fragment thereof which binds specifically to canine tumour necrosis factor alpha comprising 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 a heavy chain variable region comprising the amino acid sequence of SEQ ID NO:2 or an amino acid sequence which has an identity of at least 85% thereto, wherein the caninised antibody or antigen binding fragment thereof does not contain any amino acid in any position within the framework regions which would be foreign at that position in canines.

3. The antibody or antigen binding fragment as claimed in claim 2 wherein the antibody comprises a light chain variable region comprising the amino acid sequence of SEQ ID NO:1 and a heavy chain variable region comprising the amino acid sequence of SEQ ID NO:2.

4. The antibody or antigen binding fragment as claimed in claim 2 wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO:3 or an amino acid sequence which has an identity of at least 85% thereto and a heavy chain comprising the amino acid sequence of SEQ ID NO:5 or an amino acid sequence which has a sequence identity of at least 85% thereto.

5. The antibody or antigen binding fragment as claimed in claim 4 wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO:3 and a heavy chain comprising the amino acid sequence of SEQ ID NO:5.

6. The antibody or antigen binding fragment as claimed in claim 2 wherein the antibody comprises a light chain comprising the amino acid sequence of SEQ ID NO:8 and a heavy chain comprising the amino acid sequence of SEQ ID NO:9.

7. An antibody or an antigen binding fragment thereof which binds specifically to canine tumour necrosis factor alpha, the antibody or antigen binding fragment comprising a light chain variable region comprising:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:10, or an amino acid sequence which has a sequence identity of at least 85% thereto,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:11, or an amino acid sequence which has a sequence identity of at least 85% thereto,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:12, or an amino acid sequence which has a sequence identity of at least 85% thereto, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:13, or an amino acid sequence which has a sequence identity of at least 85% thereto,

and a heavy chain variable region comprising:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:14, or an amino acid sequence which has a sequence identity of at least 85% thereto,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:15, or an amino acid sequence which has a sequence identity of at least 85% thereto,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:16, or an amino acid sequence which has a sequence identity of at least 85% thereto, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:17, or an amino acid sequence which has a sequence identity of at least 85% thereto.

8. The antibody or antigen binding fragment thereof as claimed in claim 7, wherein the light chain variable region comprises:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:10,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:11,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:12, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:13,

and the heavy chain variable region comprises:

an FR1 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:14,

an FR2 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:15,

an FR3 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:16, and

an FR4 framework region consisting of or comprising the amino acid sequence of SEQ ID NO:17.

9. The antibody or antigen binding fragment as claimed in claim 2 or claim 7 comprising a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:4, aglycosylated SEQ ID NO:4, aglycosylated SEQ ID NO:5,

aglycosylated SEQ ID NO:6, aglycosylated SEQ ID NO:7 and SEQ ID NO:7, or an amino acid sequence which has a sequence identity of at least 85% thereto.

10. The antibody or antigen binding fragment as claimed in claim 9 wherein the antibody comprises a heavy chain comprising the amino acid sequence selected from the group consisting of SEQ ID NO:5 and SEQ ID NO:6, or an amino acid sequence which has a sequence identity of at least 85% thereto.

11. The antibody or antigen binding fragment as claimed in any one of claims 2 to 10 for use in treating, ameliorating or preventing an immune mediated condition in a canine in need thereof.

12. The antibody or antigen binding fragment as claimed in claim 11, wherein the immune mediated condition is selected from the group consisting of rheumatoid arthritis, immune mediated polyarthritis, osteoarthritis and other polyarthritidies, ankylosing spondylitis, Crohn's disease and ulcerative colitis, psoriasis, psoriatic arthritis, systemic vasculitis, atopic dermatitis, congestive heart failure, refractory uveitis, bronchial asthma and allergic conditions.

13. A pharmaceutical composition comprising a therapeutically effective amount of an antibody or an antigen binding fragment thereof as claimed in any one of claims 2 to 10 along with at least one pharmaceutically acceptable carrier, excipient or diluent.

14. Use of an antibody or antigen binding fragment as claimed in any one of claims 2 to 10 in the preparation of a medicament for the treatment, amelioration or prevention of an immune mediated condition in a canine.

15. A kit for the treatment of an immune mediated condition in a canine, the kit comprising an antibody or antigen binding fragment as claimed in any one of claims 2 to 10 and instructions for use of the same.