DKK-1 Antibodies

The present invention relates to human engineered antibodies against DKK-1 and their use in treating diseases in which pathogenesis is mediated by DKK-1.

Dickkopf-1 (Dkk-1) is a member of the dickkopf family of proteins that have been shown to be negative regulators of the canonical Wnt-signaling pathway. The pathway plays a central role in bone development and formation. Dkk-1 inhibits Wnt signaling through its interaction with the Wnt co-receptor LRP5 or LRP6 and the kremen proteins. Dkk-1 prevents members of the Wnt pathway from interacting with either LRP5 or LRP6, thus preventing Wnt-mediated signal transduction. DKK1 has also been shown to be involved in bone metastasizing cancers, including multiple myeloma, breast carcinoma, renal carcinoma and non-small cell lung cancer.

Antibodies that bind Dkk-1 have been described (for example, see WO2006015373), however, there is still a need for therapeutic human engineered DKK-1 antibodies that will inhibit the interaction of Dkk-1 with LRP5 and LRP6. In addition, in view of the involvement of Dkk-1 in the regulation of bone formation, there is a need for therapeutic human engineered anti-Dkk-1 antibodies for use in bone healing. In addition, given the involvement of Dkk-1 in cancers, there is a need for human engineered anti-Dkk-1 antibodies for treating cancers, including multiple myeloma, breast and non-small cell lung cancers.

SUMMARY

In a first aspect, there is provided a human engineered DKK-1 antibody or antigen binding fragment thereof, comprising a light chain variable region (LCVR) and a heavy chain variable region (HCVR), wherein the LCVR comprises complementarity determining regions (CDRs) LCDR1, LCDR2, and LCDR3 and the HCVR comprises CDRs HCDR1, HCDR2 and HCDR3, wherein LCDR1 has the amino sequence of SEQ ID NO:5, LCDR2 has the amino sequence of SEQ ID NO:47, LCDR3 has the amino sequence of SEQ ID NO:49, HCDR1 has the amino sequence of SEQ ID NO:1, HCDR2 has the amino sequence of SEQ ID NO:2, and an HCDR3 has the amino sequence of SEQ ID NO:44.

In a second aspect, there is provided a human engineered DKK-1 antibody comprising two light chains wherein each light chain comprises the amino acid sequence of SEQ ID NO: 20, and two heavy chains wherein each heavy chain comprises the amino acid sequence of SEQ ID NO: 18.

In a third aspect, there is provided a pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof as defined in the first aspect, and a pharmaceutically acceptable carrier, diluent, or excipient.

In a fourth aspect, there is provided the use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, as defined in the first aspect in the manufacture of a medicament for bone healing.

In a fifth aspect, there is provided the use of the human engineered DKK-1 antibody or antigen-binding fragment thereof as defined in the first aspect in the manufacture of a medicament for treating cancer.

In a sixth aspect, there is provided a pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof as defined in the second aspect, and a pharmaceutically acceptable carrier, diluent, or excipient.

In a seventh aspect, there is provided the use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, as defined in the second aspect in the manufacture of a medicament for bone healing.

In an eighth aspect, there is provided the use of the human engineered DKK-1 antibody or antigen-binding fragment thereof as defined in the second aspect in the manufacture of a medicament for treating cancer.

DETAILED DESCRIPTION

The antibodies of the present invention are therapeutically useful DKK-1 antagonists possessing a number of desirable properties. The present human engineered antibodies exhibit high affinity (Kd) to human DKK-1, cynomolgus DKK-1, rat DKK-1, mouse DKK-1, and rabbit DKK-1. Antibodies of the present invention block DKK-1-mediated inhibition of alkaline phosphatase, a marker of osteoblast activity. Furthermore, antibodies of the present invention exhibit an increase in bone mass density at both anterior and posterior cortices in a cortical defect in vivo model and demonstrate significant growth inhibition of non-small cell lung xenografts in vivo.

The present invention provides a human engineered DKK-1 antibody or antigen-binding fragment thereof that has a Kd at 37°C of less than 5.0 X 10^{-11} M for human DKK-1 (SEQ ID NO: 29), cynomolgus DKK-1 (SEQ ID NO: 30), rat Dkk-1 (SEQ ID NO: 31), mouse DKK-1 (SEQ ID NO: 33), and rabbit DKK-1 (SEQ ID NO: 32).

WE CLAIM:

1. A human engineered DKK-1 antibody or antigen binding fragment thereof, comprising a light chain variable region (LCVR) and a heavy chain variable region (HCVR), wherein the LCVR comprises the amino acid sequence of SEQ ID NO: 52 and the HCVR comprises the amino acid sequence of SEQ ID NO: 51.

2. The human engineered DKK-1 antibody of claim 1, wherein the LCVR and HCVR comprise amino acid sequences selected from the group consisting of:

   (i) a LCVR comprising the amino acid sequence of SEQ ID NO: 13 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11;

   (ii) a LCVR comprising the amino acid sequence of SEQ ID NO: 14 and a HCVR comprising the amino acid sequence of SEQ ID NO: 12;

   (iii) a LCVR comprising the amino acid sequence of SEQ ID NO: 15 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11;

   (iv) a LCVR comprising the amino acid sequence of SEQ ID NO: 16 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11.

3. The human engineered DKK-1 antibody of Claim 2, wherein the LCVR comprises the amino acid sequence of SEQ ID NO: 14 and the HCVR comprises the amino acid sequence of SEQ ID NO: 12.

4. The human engineered DKK-1 antibody of claim 1, wherein the human engineered DKK-1 antibody comprises a heavy chain and a light chain amino acid sequence selected from the group consisting of

   a. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and light chain comprising the amino acid sequence of SEQ ID NO: 19,
   b. a heavy chain comprising the amino acid sequence of SEQ ID NO: 18 and a light chain comprising the amino acid sequence of SEQ ID NO: 20,
   c. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and a light chain comprising the amino acid sequence of SEQ ID NO: 21, and

d. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and a light chain comprising the amino acid sequence of SEQ ID NO: 22.

5. The human engineered DKK-1 antibody of claim 4, comprising two light chains wherein each light chain comprises the amino acid sequence of SEQ ID NO: 20, and two heavy chains wherein each heavy chain comprises the amino acid sequence of SEQ ID NO: 18.

6. An antibody or antigen-binding fragment thereof which competes with the human engineered DKK-1 antibody or antigen-binding fragment thereof of any one of Claims 1 to 5 for binding to human DKK-1 (SEQ ID NO: 29) as determined via an antibody competition assay.

7. A pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof of any one of Claims 1 to 6, and a pharmaceutically acceptable carrier, diluent, or excipient.

8. A pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof of any one of Claims 1 to 6 together with a pharmaceutically acceptable carrier, diluent, or excipient and optionally other therapeutic ingredients.

9. A human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 for use in therapy.

10. A human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 for use in the treatment for bone healing.

11. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 in the manufacture of a medicament for bone healing.

12. A human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 for use in the treatment of cancer.

13. A human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 for use in the treatment of cancer, wherein the cancer is selected from the group consisting of multiple myeloma, breast cancer, and non-small cell lung cancer.

14. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, of any one of Claims 1 to 6 in the manufacture of a medicament for the treatment of cancer.

15. The use according to Claim 14, wherein the cancer is selected from the group consisting of multiple myeloma, breast cancer, and non-small cell lung cancer.

CLAIMS:

1. A human engineered DKK-1 antibody or antigen binding fragment thereof, comprising a light chain variable region (LCVR) and a heavy chain variable region (HCVR), wherein the LCVR comprises complementarity determining regions (CDRs) LCDR1, LCDR2, and LCDR3 and the HCVR comprises CDRs HCDR1, HCDR2 and HCDR3, wherein LCDR1 has the amino sequence of SEQ ID NO:5, LCDR2 has the amino sequence of SEQ ID NO:47, LCDR3 has the amino sequence of SEQ ID NO:49, HCDR1 has the amino sequence of SEQ ID NO:1, HCDR2 has the amino sequence of SEQ ID NO:2, and an HCDR3 has the amino sequence of SEQ ID NO:44.

2. The human engineered DKK-1 antibody of claim 1, wherein the LCVR comprises the amino acid sequence of SEQ ID NO: 52 and the HCVR comprises the amino acid sequence of SEQ ID NO: 51.

3. The human engineered DKK-1 antibody of claim 2, wherein the LCVR and HCVR comprise amino acid sequences selected from the group consisting of: (i) a LCVR comprising the amino acid sequence of SEQ ID NO: 13 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11; (ii) a LCVR comprising the amino acid sequence of SEQ ID NO: 14 and a HCVR comprising the amino acid sequence of SEQ ID NO: 12; (iii) a LCVR comprising the amino acid sequence of SEQ ID NO: 15 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11; (iv) a LCVR comprising the amino acid sequence of SEQ ID NO: 16 and a HCVR comprising the amino acid sequence of SEQ ID NO: 11.

4. The human engineered DKK-1 antibody of claim 3, wherein the LCVR comprises the amino acid sequence of SEQ ID NO: 14 and the HCVR comprises the amino acid sequence of SEQ ID NO: 12.

5. The human engineered DKK-1 antibody of claim 3, wherein the human engineered DKK-1 antibody comprises a heavy chain and a light chain amino acid sequence selected from the group consisting of a. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and light chain comprising the amino acid sequence of SEQ ID NO: 19, b. a heavy chain

comprising the amino acid sequence of SEQ ID NO: 18 and a light chain comprising the amino acid sequence of SEQ ID NO: 20, c. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and a light chain comprising the amino acid sequence of SEQ ID NO: 21, and d. a heavy chain comprising the amino acid sequence of SEQ ID NO: 17 and a light chain comprising the amino acid sequence of SEQ ID NO: 22.

6. A human engineered DKK-1 antibody comprising two light chains wherein each light chain comprises the amino acid sequence of SEQ ID NO: 20, and two heavy chains wherein each heavy chain comprises the amino acid sequence of SEQ ID NO: 18.

7. A pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof of claim 1, and a pharmaceutically acceptable carrier, diluent, or excipient.

8. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, of claim 1 in the manufacture of a medicament for bone healing.

9. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof of claim 1 in the manufacture of a medicament for treating cancer.

10. The use of claim 9, wherein the cancer is selected from the group consisting of multiple myeloma, breast cancer and non-small cell lung cancer.

11. A pharmaceutical composition comprising the human engineered DKK-1 antibody or antigen-binding fragment thereof of claim 6, and a pharmaceutically acceptable carrier, diluent, or excipient.

12. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof, of claim 6 in the manufacture of a medicament for bone healing.

13. Use of the human engineered DKK-1 antibody or antigen-binding fragment thereof of claim 6 in the manufacture of a medicament for treating cancer.

14. The use of claim 13, wherein the cancer is selected from the group consisting of multiple myeloma, breast cancer and non-small cell lung cancer.