

DESCRIPTION

Source Chinese Hamster Ovary cell line, CHO-derived
Ser30-His272
Accession # O54908

N-terminal Sequence Analysis Ser30 & Thr32

Predicted Molecular Mass 26.1 kDa

SPECIFICATIONS

SDS-PAGE 35-40 kDa, reducing conditions

Activity Measured by its ability to inhibit Wnt-3a-induced alkaline phosphatase production by MC3T3-E1 mouse preosteoblast cells. The ND_{50} for this effect is 0.02-0.1 $\mu\text{g/mL}$ in the presence of 10 ng/mL of Recombinant Mouse Wnt-3a (Catalog # 1324-WN).
Measured by its ability to inhibit Wnt induced TCF reporter activity in HEK293 human embryonic kidney cells. Recombinant Mouse Dkk-1 (Catalog # 5897-DK) inhibits a constant dose of 100 ng/mL of Recombinant Mouse Wnt-3a (Catalog # 1324-WN). The ED_{50} for this effect is 10-60 ng/mL.

Endotoxin Level <0.10 EU per 1 μg of the protein by the LAL method.

Purity >95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.

Formulation Lyophilized from a 0.2 μm filtered solution in PBS with BSA as a carrier protein. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 100 $\mu\text{g/mL}$ in PBS containing at least 0.1% human or bovine serum albumin.

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

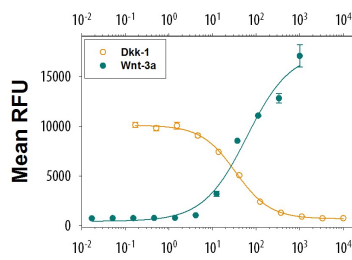
Stability & Storage Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

DATA

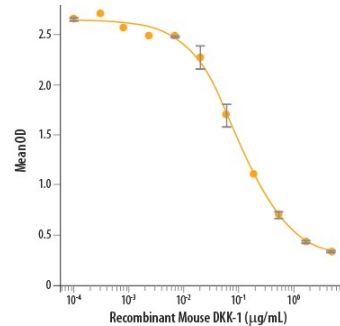
Bioactivity

Recombinant Mouse Wnt-3a (ng/mL)



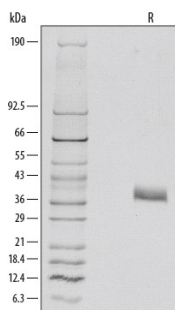
Recombinant Mouse Wnt-3a (Catalog # 1324-WN) induces a dose responsive increase in Wnt reporter activity in HEK293 cells (green circles). Recombinant Mouse Dkk-1 (Catalog # 5897-DK) inhibits a constant dose of 100 ng/mL of Recombinant Mouse Wnt-3a. The ED_{50} for this effect is 10-60 ng/mL (orange circles).

Bioactivity



Recombinant Mouse Dkk-1 (Catalog # 5439-DK) inhibits Wnt-3a-induced alkaline phosphatase production by the MC3T3-E1 mouse preosteoblast cell line. The ED_{50} for this effect is 0.02-0.1 $\mu\text{g/mL}$ in the presence of 10 ng/mL of Recombinant Mouse Wnt-3a (Catalog # 1324-WN).

SDS-PAGE



1 $\mu\text{g/lane}$ of Recombinant Mouse Dkk-1 was resolved with SDS-PAGE under reducing (R) conditions and visualized by silver staining, showing major bands at 35-40 kDa. Multiple bands in gel are due to variable glycosylation.

BACKGROUND

Dickkopf related protein 1 (Dkk-1) is the founding member of the Dickkopf family of proteins that includes Dkk-1, -2, -3, -4, and a related protein, Soggy (1, 2). Dkk proteins are secreted proteins that contain two conserved cysteine-rich domains separated by a linker region. Each domain contains ten cysteine residues (1-3). Mature mouse Dkk-1 is a 40 kDa glycosylated protein that shares 86%, 96%, 83% and 82% amino acid (aa) sequence identity with human, rat, rabbit and bovine Dkk-1, respectively. It also shares 41% and 36% aa identity with human Dkk-2 and Dkk-4, respectively. Dkk-1 and Dkk-4 are well documented antagonists of the canonical Wnt signaling pathway (1, 2). This pathway is activated by Wnt engagement of a receptor complex composed of the Frizzled proteins and one of two low-density lipoprotein receptor-related proteins, LRP5 or LRP6 (4). Dkk-1 antagonizes Wnt by forming ternary complexes of LRP5/6 with Kremen1 or Kremen2 (4, 5). Dkk-1/LRP6/Krm2 complex internalization has been shown to down-regulate Wnt signaling (4, 5). Dkk-1 is expressed throughout development and antagonizes Wnt-7a during limb development (6, 7). Other sites of expression include developing neurons, hair follicles and the retina of the eye (8, 9). The balance between Wnt signaling and Dkk-1 inhibition is critical for bone formation and homeostasis (10). Insufficient or excess Dkk-1 activity in bone results in increased or decreased bone density, respectively (8, 11). In adults, Dkk-1 is expressed in osteoblasts and osteocytes, and neurons. Cerebral ischemia induces Dkk-1 expression, which contributes to neuronal cell death (12).

References:

1. Glinka, A. *et al.* (1998) *Nature* **391**:357.
2. Niehrs, C. (2006) *Oncogene* **25**:7469.
3. Bullock, C.M. *et al.* (2004) *Mol. Pharmacol.* **65**:582.
4. Mao, B. *et al.* (2001) *Nature* **411**:321.
5. Mao, B. *et al.* (2002) *Nature* **417**:664.
6. Kemp, C. *et al.* (2005) *Dev. Dyn.* **233**:1064.
7. Adamska, M. *et al.* (2004) *Dev. Biol.* **272**:134.
8. Li, J. *et al.* (2006) *Bone* **36**:754.
9. Verani, R. *et al.* (2006) *J. Neurochem.* **101**:242.
10. Pinzone, J.J. *et al.* (2009) *Blood* **113**:517.
11. Morvan, F. *et al.* (2006) *J. Bone Miner. Res.* **21**:934.
12. Cappuccio, I. *et al.* (2005) *J. Neurosci.* **25**:2647.