Recombinant Human BMP-4

Catalog Number: 314E-GMP

DESCRIPTION

Source	*E. coli*-derived human BMP-4 protein Lys303-Arg408 Accession # P12644.1 Produced in an animal-free laboratory. Manufactured and tested under cGMP guidelines
N-terminal Sequence Analysis	Identity was confirmed by mass spectrometry
Structure / Form	Disulfide-linked homodimer
Predicted Molecular Mass	12 kDa

SPECIFICATIONS

SDS-PAGE	8-9 kDa, reducing conditions
Activity	Measured by its ability to induce BMP responsive SEAP reporter activity in HEK293 human embryonic kidney cells. The ED_{50} for this effect is 3-21 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.
Host Cell Protein	<0.5 ng per µg of protein when tested by ELISA.
Mycoplasma	Negative for Mycoplasma.
Host Cell DNA	<0.0015 ng per µg of protein when tested by PCR.
Formulation	Lyophilized from a 0.2 µm filtered solution in Acetonitrile and TFA. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 50-200 µg/mL in sterile 4 mM HCl.
Shipping	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • A minimum of 12 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 3 months, ≤ -65 °C under sterile conditions after reconstitution.

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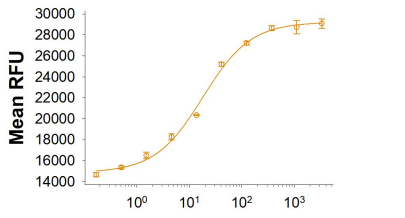
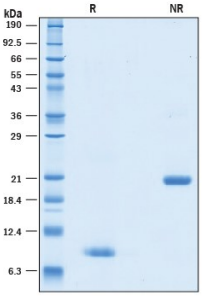
DATA

Bioactivity



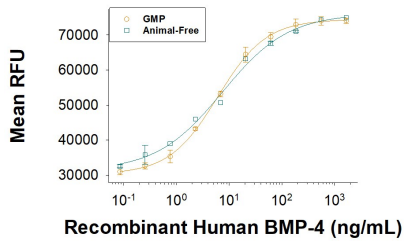
GMP-grade Recombinant Human BMP-4 (Catalog # 314E-GMP) induces BMP responsive SEAP reporter activity in HEK293 human embryonic kidney cells. The ED_{50} for this effect is 3-21 ng/mL.

SDS-PAGE



2 µg/lane of GMP-grade Recombinant Human BMP-4 Protein (Catalog # 314E-GMP) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 8-9 kDa and 20-22 kDa, respectively.

Bioactivity



Equivalent Bioactivity of GMP and Animal-Free grades of Recombinant Human BMP-4
Equivalent bioactivity of GMP (Catalog # 314E-GMP) and Animal-Free (Catalog # AFL314E) grades of Recombinant Human BMP-4 as measured by its ability to induce BMP responsive SEAP reporter activity in HEK293 human embryonic kidney cells (orange and green respectively).

BACKGROUND

Bone morphogenetic protein 4 (BMP-4) is a TGF-beta superfamily ligand that is widely expressed from early embryogenesis through adulthood. It plays an important role in mesenchyme formation, epidermal determination, suppression of neural induction, the development of multiple organs, and tissue repair (1-5). It is an integral part of many stem cell differentiation pathways, including lung tissue, (6), adipogenesis (7) and osteogenesis (8, 9). The human BMP-4 precursor contains a 273 amino acid (aa) propeptide and a 116 amino acid (aa) mature protein (10). Processing of the propeptide by furin or proprotein convertase 6 enables the formation of the mature disulfide-linked homodimeric BMP-4 and facilitates its secretion. Similar intracellular processes may lead to the formation and recreation of BMP4/BMP7 disulfide-linked heterodimer (11-13). Mature human and mouse BMP-4 share 98% aa sequence identity. Human BMP-4 shares 85% aa sequence identity with human BMP-2 and less than 50% with other human BMPs. In Xenopus, BMP-4 dimers provide ventralizing signals for existing mesoderm (14). BMP-4 signals through tetrameric complexes composed of type I (primarily Activin RIA or BMPR-IA) and type II (primarily Activin RIIA or BMPR-II) receptors (15, 16). The bioavailability of BMP-4 is regulated by its interaction with multiple proteins and glycosaminoglycans (17-19).

References:

1. Zhang, P. *et al.* (2008) Blood **111**:1933.
2. Gambaro, K. *et al.* (2006) Cell Death Differ. **13**:1075.
3. Simic, P. and S. Vukicevic (2005) Cytokine Growth Factor Rev. **16**:299.
4. Sadlon, T.J. *et al.* (2004) Stem Cells **22**:457.
5. Frank, D.B. *et al.* (2005) Circ. Res. **97**:496.
6. Lee, J-H. *et al.* (2014) Cell. **156**:440.
7. Tang, QQ. *et* Lane MD. (2012) Annu Rev. Biochem. **81**:715.
8. Urist, MR. (1965). Science. **150**:893.
9. Bandyopadhyay, A. *et al.* (2006) Plos Genetics. 2:e216.
10. Wozney, J. *et al.* (1988) Science **242**:1528.
11. Cui, Y. *et al.* (1998) EMBO J. **17**:4735.
12. Cui, Y. *et al.* (2001) Genes Dev. **15**:2797.
13. Aono, A. *et al.* (1995) Biochem. Biophys. Res. Commun. **210**:670.
14. Nishimatsu, S. and G.H. Thomsen (1998) Mech. Dev. **74**:75.
15. Chen, D. *et al.* (2004) Growth Factors **22**:233.
16. Lavery, K. *et al.* (2008) J. Biol. Chem. April 24 epub.
17. Rosen, V. (2006) Ann. N.Y. Acad. Sci. **1068**:19.
18. Jones, C.M. and J.C. Smith (1998) Dev. Biol. **194**:12.
19. Takada, T. *et al.* (2003) J. Biol. Chem. **278**:43229.

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