Recombinant Human Wnt-3a

Catalog Number: 5036-WN/CF

DESCRIPTION

Source	Chinese Hamster Ovary cell line, CHO-derived human Wnt-3a protein Ser19-Lys352 Accession # NP_149122
N-terminal Sequence Analysis	Ser19
Predicted Molecular Mass	37.4 kDa

SPECIFICATIONS

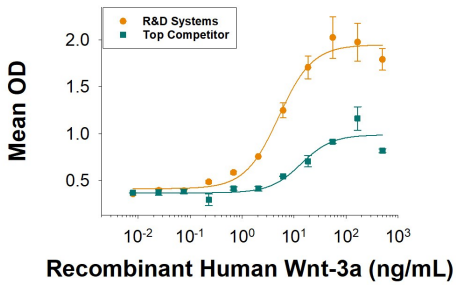
SDS-PAGE	40 kDa, reducing conditions
Activity	Measured by its ability to induce alkaline phosphatase production by MC3T3-E1 mouse preosteoblast cells. The ED_{50} for this effect is 5-25 ng/mL. Measured by its ability to induce Topflash reporter activity in HEK293T human embryonic kidney cells. The ED_{50} for this effect is <500 ng/mL. Protein concentrations should be titrated based on cell type and if appropriate, passage number of the cell line. **Optimal concentrations should be determined by each laboratory for each application.**
Endotoxin Level	<1.0 EU per 1 µg of the protein by the LAL method.
Purity	>75%, by SDS-PAGE under reducing conditions and visualized by silver stain.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS, EDTA and CHAPS. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 200 µg/mL in sterile PBS.
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 Human Wnt-3a (Catalog # 5036-WN/CF) induces alkaline phosphatase production by the MC3T3-E1 mouse preosteoblast cell line. The ED_{50} is 1.7-fold better with more than twice the maximum response compared to the top competitor's Wnt-3a.

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BACKGROUND

Wnt-3a is one of 19 vertebrate members of the Wingless-type MMTV integration site (Wnt) family of highly conserved cysteine-rich secreted glycoproteins important for normal developmental processes (1). Wnts bind to the cell surface Frizzled family receptors in conjunction with low-density lipoprotein receptor-related protein family receptors (LRP5 or 6) resulting in the stabilization of intracellular β-catenin levels (2). As intracellular β-catenin levels rise, β-catenin binds to TCF/LEF transcription factors leading to expression of Wnt target genes (3). Endo-IWR 1 (Catalog # 3532, # PSM1324) is a cell-permeant small molecule inhibitor of Axin turnover that suppresses Wnt signal transduction by stabilizing the β-catenin destruction complex (4). Wnt-3a is a 44 kDa secreted hydrophobic glycoprotein containing a conserved pattern of 24 cysteine residues (5). Wnt-3a has two N-linked glycosylation sites (Asn 87, Asn 298), and Ser 209 is modified with palmitoleic acid (6). Glycosylation and acylation are essential for efficient Wnt secretion and biological activity, respectively (6, 7). Human Wnt-3a shares 96% amino acid (aa) identity with mouse mouse, bovine and canine Wnt-3a, and 89%, 86% and 84% aa identity with chicken, Xenopus and zebrafish Wnt-3a, respectively. It also shares 87% aa identity with Wnt3. During embryonic development, Wnt-3a is necessary for proper development of the hippocampus, anterior-posterior patterning, somite development, and tailbud formation (9-12). Wnt-3a also promotes self-renewal of hematopoietic stem cells, neural stem cells, and embryonic stem cells (13, 14).

References:

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