Recombinant Human Dkk-3

Catalog Number: 1118-DK

DESCRIPTION

Source	Mouse myeloma cell line, NS0-derived human Dkk-3 protein Ala22-Ile350, with a C-terminal 10-His tag Accession # Q9UBP4.1
N-terminal Sequence Analysis	Ala22
Predicted Molecular Mass	37.5 kDa

SPECIFICATIONS

SDS-PAGE	64-74 kDa, reducing conditions
Activity	Measured by its ability to inhibit proliferation of HeLa human cervical epithelial carcinoma cells. Hsieh, S.Y. *et al.* (2004) Oncogene **23**:9183. The ED_{50} for this effect is 3-12 µg/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. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 250 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.

BACKGROUND

Dkk-3, also known as REIC (Reduced Expansion in Immortalized Cells), is one of four numbered members of the Dickkopf family of Wnt antagonists (1). Dkk-3 is a secreted monomer expressed in many normal human tissues, most strongly in heart, brain and spinal cord (1, 2), and during early embryonic development in the mouse (3). N-glycosylation at up to four sites preceding or between two conserved cysteine-rich motifs results in expression of a 45 - 65 kDa glycoprotein (1, 4). The cysteine-rich motifs contain 10 cysteines each, with prokineticin and colipase families containing sequences similar to those of the second motif (1, 5). Human Dkk-3 shows 82%, 88%, 85% and 53% amino acid (aa) identity with mouse, bovine, canine and chick Dkk-3, respectively, and 37 - 45% aa identity with other human Dkk family members. Several lines of evidence implicate Dkk-3 as a negative growth regulator. Dkk-3 is downregulated in many tumors as compared to normal cells, sometimes by loss of heterozygosity (4, 6). Downregulation by CpG hypermethylation in acute lymphoblastic leukemia is correlated with faster progression and shorter survival (7). Release of cultured cells from serum starvation results in downregulation of Dkk-3 in late G1 phase of the cell cycle (6). Overexpression of Dkk-3 results in tumor cell-line-specific growth inhibition, induction of apoptosis, and decreased tumorigenicity in nude mice (2, 4, 6). The prototype Dickkopf member, Dkk-1, antagonizes Wnt family signaling by binding to Wnt receptors LRP5 and LRP6 (low-density lipoprotein receptor-related proteins) and promoting their internalization (1, 9, 10). Results are less straightforward for Dkk-3, where some studies show binding to LRP5/6 while others do not. These effects appear to be dependent on the cells and conditions used (1, 6 - 10).

References:

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