

**DESCRIPTION**

<b>Source</b>	Mouse myeloma cell line, NS0-derived		
	Human Frizzled-4 (Phe37-Glu180) Accession # Q9ULV1	IEGRMD	Human IgG <sub>1</sub> (Pro100-Lys330)
	N-terminus		C-terminus
<b>N-terminal Sequence Analysis</b>	Phe37		
<b>Structure / Form</b>	Disulfide-linked homodimer		
<b>Predicted Molecular Mass</b>	42.8 kDa (monomer)		

**SPECIFICATIONS**

<b>SDS-PAGE</b>	58-60 kDa, reducing conditions
<b>Activity</b>	Measured by its binding ability in a functional ELISA. In a 100 µL reaction mixture containing biotinylated rmWnt-5a at 100 ng/mL and rhFrizzled-4/Fc Chimera dilutions at 0.1-2,000 ng/mL, the concentration of rhFrizzled-4/Fc Chimera that produces 50% of the optimal binding response is found to be approximately 15-60 ng/mL. <b>Optimal concentrations should be determined by each laboratory for each application.</b>
<b>Endotoxin Level</b>	<0.10 EU per 1 µg of the protein by the LAL method.
<b>Purity</b>	>95%, by SDS-PAGE under reducing conditions and visualized by silver stain.
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 400 µg/mL in PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>● 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>● 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>● 3 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

**BACKGROUND**

Frizzled-4, designated CD344, is a 7-transmembrane glycoprotein of the Frizzled family within the G-protein coupled receptor superfamily (1, 2). Frizzled proteins function as receptors for Wnt proteins and can activate canonical Wnt/beta-catenin signaling as well as planar cell polarity and calcium flux pathways (1). Frizzled-4 is particularly important in angiogenic Wnt pathway signaling (1, 5). Frizzleds contain a divergent N-terminal signal peptide, a highly conserved ~120 amino acid (aa) cysteine-rich domain (CRD), a variable length linker region, seven transmembrane domains, and a variable-length C-terminal tail (1). The human Frizzled-4 cDNA encodes 537 aa with a 36 aa signal sequence and a 186 aa N-terminal extracellular sequence (4). The portion expressed in this product includes aa 37-180, and shares 93%, 93%, 94%, 95% and 97% identity with the corresponding region of mouse, rat, bovine, canine and equine Frizzled-4, respectively. This portion competes for Wnt binding with endogenous receptors. A human 122 aa soluble form that diverges at aa 95 is proposed to be a positive regulator of Wnt signaling pathways (5). Frizzled-4 is unusual in binding a non-wnt ligand, Norrin, in addition to binding Wnt ligands (1, 3, 6). Norrin binds the Frizzled-4 CRD, activates Wnt signaling pathways and uses LRP5/6 as co-receptors (3, 6). Deletion of either Frizzled-4 or Norrin in mice results in a similar phenotype including malformation of vasculature in the retina, cerebellar degeneration, and loss of hair cells in the inner ear (1, 3, 7). In humans, blindness due to familial exudative vitreoretinopathy (FEVR) is associated with mutations producing loss of function of Frizzled-4 or Norrin, designated EVR1 and EVR2, respectively (1, 3, 8). Frizzled-4 expression has been found in many tissues, including mouse ovary, where it influences corpus luteum vasculogenesis and is necessary for fertility (9).

**References:**

1. Huang, H-C. and P.S. Klein (2004) *Genome Biol.* **5**:234.
2. Parmalee, N.L. and J. Kitajewski (2008) *Curr. Drug Targets* **9**:558.
3. Xu, Q. *et al.* (2004) *Cell* **116**:883.
4. Kirikoshi, H. *et al.* (1999) *Biochem. Biophys. Res. Commun.* **264**:955.
5. Sagara, N. *et al.* (2001) *Biochem. Biophys. Res. Commun.* **282**:750.
6. Smallwood, P.M. *et al.* (2007) *J. Biol. Chem.* **282**:4057.
7. Wang, Y. *et al.* (2001) *J. Neurosci.* **21**:4761.
8. Robitaille, J. *et al.* (2002) *Nat. Genet.* **32**:326.
9. Hsieh, M. *et al.* (2005) *Biol. Reprod.* **73**:1135.