

## DESCRIPTION

<b>Species Reactivity</b>	Human/Mouse
<b>Specificity</b>	Detects human and mouse Frizzled-4 in Western blots. In Western blots, no cross-reactivity with recombinant mouse (rm) Frizzled-1, -2, -3, -6, -7, -8, or recombinant human Frizzled-5 is observed. Detects Frizzled-4 by flow cytometry and immunohistochemistry on mouse differentiated embryoid bodies derived from the mouse embryonic cell line, D3.
<b>Source</b>	Monoclonal Rat IgG <sub>2A</sub> Clone # 145901
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	Mouse myeloma cell line NS0-derived recombinant mouse Frizzled-4 Phe37-Glu180 Accession # Q61088
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

## APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
<b>Western Blot</b>	1 µg/mL	Recombinant Human Frizzled-4 (Catalog # 5847-FZ) Recombinant Mouse Frizzled-4 Fc Chimera (Catalog # 194-FZ)
<b>Flow Cytometry</b>	2.5 µg/10 <sup>6</sup> cells	HEK293 human embryonic kidney cell line
<b>Immunocytochemistry</b>	8-25 µg/mL	Immersion fixed HEK293 human embryonic kidney cell line
<b>CyTOF-ready</b>	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <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>● 6 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 mouse Frizzled-4 cDNA encodes 537 aa with a 36 aa signal sequence and a 186 aa N-terminal extracellular sequence (4). The portion expressed includes aa 37-180, and shares 93%, 94%, 90%, 89% and 88% identity with human, rat, equine, bovine and canine Frizzled-4, respectively. This portion competes for Wnt binding with endogenous receptors. In humans, a 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 (4, 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. Wang, Y. *et al.* (1996) *J. Biol. Chem.* **271**:4468.
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.