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# ProductInformation

Flt-3/Fc Chimera Mouse, Recombinant Expressed in NSO cells

Product Number F 2803

## **Product Description**

Recombinant Mouse FIt-3/Fc Chimera<sup>1</sup> is produced from a DNA sequence encoding the signal sequence of human IL-3 Receptor  $\alpha$  (Met 1 - Gln 18) linked to the extracellular domain of mouse FIt-3 (Asn 28 - Ser 544) fused to the C-terminal histidine-tagged Fc region of human IgG1 by a linker peptide. The chimeric protein is expressed in mouse myeloma NSO cells. Based on N-terminal sequencing, recombinant mouse FIt-3, a disulfide-linked homodimer, has Asn 28 as the aminoterminus. Mouse FIt-3/Fc contains 760 amino acids with a calculated mass of approximately 85 kDa. As a result of glycosylation, mouse FIt-3/Fc monomer migrates as a 116 kDa protein in SDS-PAGE. At the amino acid level, human and mouse FIt-3 are approximately 85% identical.

Flt-3 (*fms*-like tyrosine kinase-3) receptor, also known as Flk-2 (fetal liver kinase) and Stk-1 (stem cell tyrosine kinase), is a member of the class III subfamily of receptor tyrosine kinases.<sup>1-4</sup> Additional members of this receptor family are the receptors for macrophage-colony-stimulating factor and steel factor, encoded by the *KIT*<sup>5,6</sup> and *FMS*<sup>7,8</sup> protooncogenes, respectively, and the receptors for  $\alpha$ - and  $\beta$ -platelet -derived growth factors (PDGFRA and -B). Common structural features include the extracellular region composed of five immunoglobulin-like domains and an intracellular tyrosine kinase made up of an ATP-binding loop and a catalytic domain separated by a kinase insert domain.

Flt-3, Fms, and Kit play a key role in hematopoiesis by stimulating proliferation and/or differentiation of various hematopoietic cell types.<sup>9, 10</sup> Mice lacking a functional Flt-3 receptor have normal mature hematopoietic populations; however, they exhibit reduced numbers of early B cell precursors and multipotent stem cells.<sup>11</sup>

Recombinant soluble Flt-3/Fc chimera binds FL (Flt-3 ligand) with high affinity and is a potent FL antagonist. Flt-3 ligand (FL) is a transmembrane protein with structural homology to macrophage colony stimulating factor (M-CSF) and stem cell factor (SCF) that promotes growth of early B cell progenitor cells and induces adhesion of the precursor B cell line BaF3/Fkt3 to fibronectin.

The Flt-3 receptor is expressed in a variety of tissues including placenta, gonads, and tissues of nervous and hematopoietic origin. In the hematopoietic system, the expression of Flt-3/Flk-2 ligand and Flt-3 receptor is restricted to the enriched stem/progenitor cells.<sup>2</sup>

#### Reagent

Recombinant Mouse FIt-3/Fc Chimera is supplied as approximately 50  $\mu$ g of protein lyophilized from a 0.2  $\mu$ m filtered solution in phosphate buffered saline (PBS).

#### **Preparation Instructions**

Reconstitute the contents of the vial using 0.2  $\mu$ m filtered phosphate buffered saline containing at least 0.1% human serum albumin or bovine serum albumin. Prepare a stock solution of no less than 10  $\mu$ g/ml.

## Storage/Stability

Store at -20 °C. Upon reconstitution, the product may be stored at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. Do not store in a "frost-free" freezer.

## **Product Profile**

Recombinant Mouse Flt-3/Fc Chimera is measured by its ability to inhibit recombinant mouse Flt-3 ligandinduced proliferation of a Flt-3 transfected pro-B cell line.

The ED<sub>50</sub> for this effect is typically 0.05 to 0.25  $\mu$ g/ml in the presence of recombinant mouse Flt-3 ligand (25 ng/ml).

The  $ED_{50}$  is defined as the effective concentration of growth factor that elicits a 50% increase in cell growth in a cell based bioassay.

Purity: >95 % as determined by SDS-PAGE, visualized by silver stain.

Endotoxin level is < 0.1 ng/ $\mu$ g cytokine as determined by the LAL (Limulus amebocyte lysate) method.

#### References

- 1. Matthews, W., Cell, **65**, 1143-1152 (1991).
- Small, D., et al., Proc. Natl. Acad. Sci. USA, 91, 459-463 (1994).
- Rosnet, O., and Birnbaum, D., Crit. Rev. Oncog. 4, 595-613 (1993).
- 4. Rosnet, O., et al., D., Blood, 82, 1110-1119 (1993).
- 5. Chabot, B., et al., Nature, **335**, 88-89 (1988).
- 6. Geissler, E.N., et al., Cell, **55**, 185-192 (1988).
- 7. Woolford, J., et al., Cell, **55**, 965-977 (1988).
- Rothwell, V.M., and Rohrschneider, L. R., Oncogene Res., 1, 311-324 (1987).
- Rohrschneider, L.R. in Guidebook to Cytokines and Their Receptors, (Nicola, A., ed), Oxford University Press, Oxford, UK, pp. 168-170 (1995).
- Lyman, S.D., and Jacobsen, S.E., Blood, **91**, 1101-1134 (1998).
- 11. Mackarehtschian, K., et al., Immunity, **3**, 147-161 (1995).

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