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# **ProductInformation**

# Osteoprotegerin/Fc Chimera.

Mouse, Recombinant Expressed in mouse NSO cells

Product Number O 8137

Synonyms: OPG, OCIF, Osteoclastogenisis Inhibitory Factor, TNFRSF11B (Tumor Necrosis Factor Receptor Superfamily Member 11B)

## **Product Description**

Recombinant mouse osteoprotegerin/Fc chimera consists of a human IL-3 R $\alpha$  sequence (Met 1-Gln 18) fused to the N-terminal end of amino acid residues Glu 22-Leu 401 of mouse OPG. The OPG portion was then fused by means of a polypeptide linker to the Fc portion of human IgG1 that is 6X histidine-tagged at the carboxyl terminus. The chimeric protein is expressed in a mouse myeloma cell line, NSO. Recombinant OPG is a disulfide-linked homodimer. The amino terminus is Glu(22) based on N-terminal sequencing. The calculated molecular mass of the reduced OPG/Fc monomer is approximately 70.9 kDa, but as a result of glycosylation, the recombinant OPG/Fc chimera migrates as an approximately 100 kDa protein in SDS-PAGE.  $^1$ 

Osteoprotegerin is a soluble secreted protein that posesses no apparent cell-associated motifs. It is also referred to as TNFRSF11B (TNF receptor superfamily, member 11B). OPG was originally isolated by sequence homology as a TNF receptor family protein in fetal rat intestine. OCIF, initially believed to be a unique cytokine, was isolated from human embryonic fibroblasts. Subsequent studies have shown that the OPG gene maps to chromosome 8q23-24 and is identical to OCIG. The amino-terminal half of OPG contains four cysteine-rich repeats characteristic of TNF receptor family members. The carboxy-terminal of OPG was found to contain two death domain homologous regions in tandem. Human and mouse OPG share approximately 84% and 94% homology with rat OPG. Native OPG has been found to exist primarily as disulfide-linked dimers. Two TNF superfamily ligands, OPG ligand/TRANCE/ODF/RANKL and TRAIL/APO-2, have been shown to be cellular ligands for OPG. Mouse OPG transcripts are expressed in liver, lung, heart, and kidney tissue. OPG mRNA is expressed at high levels in stomach, intestines, skin, and calvaria. In humans, high levels are detected in the lung, heart, kidney, and placenta.

## Reagents

Recombinant mouse OPG/Fc chimera is supplied as approximately 100  $\mu$ g of protein lyophilized from sterile filtered phosphate-buffered saline (PBS).

# **Preparation Instructions**

Reconstitute the vial contents with sterile PBS containing 0.1% HSA or BSA. Stock solution concentration should be no less than  $100 \mu g/ml$ .

# Storage/Stability

Lyophilized samples are stable for more than six months at  $-20~^{\circ}$ C. Upon reconstitution, store at 2-8  $^{\circ}$ C for up to one month. For extended storage, store in working aliquots at  $-20~^{\circ}$ C. Repeated freeze-thaw cycles should be avoided. Do not store in a frost-free freezer.

### **Product Profile**

OPG/Fc activity is measured by its ability to neutralize apoptosis in L-929 cells treated with 50 ng/ml of rhTRAIL. The ED $_{50}$  range for this assay is typically 25 – 50 ng/ml. In the presence of 20 ng/ml cross-linked rhTRAIL, the ED $_{50}$  shifts to 8 – 15 ng/ml. Optimal dilutions should be determined by each laboratory for each application.

Purity: >95% (SDS-PAGE, visualized by silver stain)

Endotoxin level: < 0.1 ng/µg of protein as determined by the LAL (Limulus amebocyte lysate) method.

#### References

 Simonet, W.S., et al., Osteoprotegrin: A Novel Secreted Protein Involved in the Regulation of Bone Density, Cell, 89:309-319 (1997).

- 2. Lacey, D.L., et al., Osteoprotegrin Ligand Is a Cytokine that Regulates Osteoclast Differentiation and Activation, Cell, 93:165-176 (1998).
- 3. Emery, J.G., et al., Osteoprotegrin is a Receptor for the Cytotoxic Ligand TRAIL, J. Biol. Chem., 273:14363-14367 (1998).

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