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ProductInformation

Osteopontin
Human, Recombinant
Expressed in NSO cells

Product Number **O 4264**

Product Description

Recombinant Human Osteopontin (OPN) is produced from a DNA sequence encoding the signal peptide from human IL-3 Rα fused to the C-terminal 6X histidine-tagged mature human Osteopontin, expressed in mouse myeloma NSO cells.[1] The mature protein was produced by the proteolytic removal of the signal peptide. From N-terminal sequencing, the protein starts at Ile 17 and has a calculated molecular mass of 32.9 kDa. As a result of glycosylation, recombinant human OPN migrates as a 60-65 kDa protein in SDS-PAGE under reducing conditions. Human, mouse, rat, pig, and bovine osteopontin share approximately 40% amino acid sequence identity.

Human OPN encodes a 314 amino acid residue precursor protein with a 16 amino acid residue predicted signal peptide that is cleaved to yield a 298 amino acid residue mature protein with an integrin binding sequence (RGD), and N- and O-glycosylation sites. By alternative splicing, there are at least three human OPN isoforms.

Osteopontin (OPN, also known as transformation-associated secreted phosphoprotein, bone sialoprotein-1, 2ar, 2B7, early T lymphocyte activation protein-1 (ETA-1), minopotin, and calcium oxalate crystal growth inhibitor protein) is a secreted acidic phosphorylated glycoprotein. Osteopontin has important functions in bone metabolism and inflammatory processes. OPN binds various cell types through RGD-mediated interaction with the integrins $\alpha_v\beta_1$, $\alpha_v\beta_3$, $\alpha_v\beta_5$, and non-RGF-mediated interactions with CD44 variants and integrins ($\alpha_8\beta_1$ or $\alpha_9\beta_1$).[2]

Osteopontin (OPN), originally isolated from bone matrix, is also found in kidney, placenta, blood vessels, and various tumor tissues. Many cell types (macrophages, osteoclasts, activated T-cells,[3] fibroblasts, epithelial cells, vascular smooth muscle cells, and natural killer cells) express osteopontin in response to activation by cytokines, growth factors, or inflammatory mediators. Increased expression of OPN is associated with numerous conditions such as atheroschlerotic plaques, renal tubulointerstitial fibrosis, granuloma formations in tuberculosis and silicosis,[4] neointimal formation associated with balloon catheterization, metastasizing tumors, and cerebral ischemia. OPN is chemotactic for macrophages, smooth muscle cells, endothelial cells, and glial cells. OPN inhibits nitric oxide production and cytotoxicity by activated macrophages. In various cell types, OPN can be used as a substrate for proteolytic cleavage by thrombin, enterokinase, MMP-3, and MMP-7.[5, 6]

Reagent

Recombinant Human Osteopontin is supplied as approximately 50 µg of protein lyophilized from a 0.2 µm filtered solution of phosphate buffered saline containing 2.5 mg bovine serum albumin.

Storage/Stability

Prior to reconstitution, store at –20 °C. Reconstituted product should be stored at –20 °C in working aliquots. Avoid repeated freezing and thawing. Do not store in a frost-free freezer.

Preparation Instructions

Reconstitute the contents of the vial using 0.2 µm filtered phosphate buffered saline containing 0.1% human serum albumin or bovine serum albumin. Prepare a stock solution of no less than 10 µg/ml.

Product Profile

The biological activity of recombinant human osteopontin is measured by its ability to mediate 293 cell adhesion.

Recombinanat Human Osteopontin immobilized at 1 μg/ml (100 μl/well) mediates adhesion of added 293 cells >30% (100 μl/well at 10^6 cells/ml).

Human thrombin treatment will increase 293 adhesion by approximately 5- fold.[5]

References

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