

Osteopontin

Osteopontin (OPN) is secreted by both osteoblasts and osteoclasts and has been shown to affect the bone remodeling process by differentiating osteoclasts, inhibiting mineral deposits, and enhancing osteoclast activity.¹ High levels of OPN can be found along bone cement lines and in locations where osteoclasts are interfacing with new bone cells (laminae limitantes).¹ Once secreted and cleaved, OPN binds with integrin and CD44 receptors. As such, OPN plays a vital role in regulating bone resorption.

Featured Assays

ALPCO offers efficient and robust osteopontin assays for human and mouse testing. Results are achieved in three hours with $\leq 10 \mu\text{L}$ of sample and the assays employ a seven point calibration curve with the inclusion of a native plasma control for added confidence.

Human Osteopontin ELISA

Catalog #:	41-OPNHU-E01
Sample Type:	Heparin Plasma, Urine
Sample Size:	10 μL
Range:	0.31 - 20 ng/mL
Sensitivity:	0.05 ng/mL
Incubation:	3 hours

Mouse Osteopontin ELISA

Catalog #:	41-OPNMS-E01
Sample Type:	Heparin Plasma, Urine
Sample Size:	5 μL
Range:	0.031 - 2 ng/mL
Sensitivity:	0.015 ng/mL
Incubation:	3 hours

Assay Procedure

1. Add 100 μL of standards, control, or diluted samples
2. Incubate for 2 hours at RT
3. Wash 4 times
4. Add 100 μL biotin conjugate
5. Incubate 20 minutes in the dark at RT
6. Wash 4 times
7. Add 100 μL HRP-streptavidin conjugate
8. Incubate 20 minutes in the dark at RT
9. Wash 4 times
10. Add 100 μL TMB substrate
11. Incubate 10 minutes in the dark at RT
12. Add 100 μL stop solution and read at 450 nm

Assays are For Research Use Only

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Osteopontin

Osteopontin (OPN) is a highly diverse protein that is a member of the Small Integrin-Binding Ligand, N-linked Glycoprotein (SIBLING) family of proteins which has been mapped to human chromosome 4.² OPN consists of about 300 amino acids and is a highly acidic protein. It contains an arginine-glycine-aspartic acid (RGD)-binding site, two heparin-binding sites, one thrombin cleavage site, and a putative calcium binding site.¹ The protein is highly phosphorylated (serine/threonine) and glycosylated. Due to the extensive post-translational modifications (PTM), the protein can vary in size from 32 to 75 kDa.

OPN is involved in many biological processes including, but not limited to:

- Bone remodeling
- Pathogenesis of atherosclerosis
- Cancer
- Chronic inflammatory disease
- Deregulation of apoptosis
- Chemotaxis

Most of these processes are dependent upon the phosphorylation state of OPN and cleavage status. As cells secrete OPN into the extracellular matrix, the relatively bio-inactive full length OPN is rapidly cleaved through protease activity, which results in two functionally active proteins (N-terminal sequence which interacts with integrin protein receptors and the C-terminal sequence which interacts with CD44 receptors). Multiple proteases perform this cleavage; however, thrombin most often performs the cleavage.¹

Synthesis and secretion of OPN occurs in many tissues including, but not limited to:

- Osteoblasts
- Osteocytes
- Endothelial cells
- Skeletal muscle myoblasts
- Smooth muscle
- Bone marrow cells

In human adults however, OPN is secreted into bodily fluids (blood, plasma, urine, breast milk) by specific cells found in bone, epithelial tissues, and the kidneys. The regulatory mechanism of OPN is not completely understood but it has been demonstrated that OPN regulation differs depending on cell type and specific function.

Areas Of Interest

Cancer

OPN overexpression has been shown to correlate with tumor progression, poor prognosis, and increased invasiveness.³ Through the interaction with integrin and CD44 receptors on cell surfaces, OPN can activate various intracellular pathways that are known to enhance cell adhesion, migration, vascularization, angiogenesis, and apoptosis resistance. The interest of OPN as a potential cancer biomarker, or pharmaceutical therapeutic target, has increased as more studies have expanded upon the role of OPN in tumorigenesis and cancer cell progression.

Immune Response

OPN acts as a key immune system regulator, particularly with regards to macrophage activity/differentiation, T-cell activity/maturation, and dendritic cell response/chemotaxis. During Macrophage differentiation, OPN secretion is significantly increased in response to various inflammatory cytokines, e.g. IFN- γ , TNF- α , IL-6, IL-1 β . These increased levels of OPN affect macrophage function and bio-activity. OPN acts as a chemotactic agent by promoting the migration of microphages to the site of acute/chronic inflammation. As such, macrophages have demonstrated to be a significant target and source of OPN.³ OPN has become an increasingly important biomarker as the relationship between OPN and the immune response becomes better understood.

References

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