**OSTEOPONTIN: A DIVERGENT PROTEIN MOLECULE**

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**ABSTRACT**

Osteopontin (OPN) is a phosphoglycoprotein with a multidomain structure and functions characteristic of a matricellular protein. OPN interacts with cell surface receptors via arginine-glycine-aspartate (RGD) and other multiple non-RGD containing adhesive domains. OPN has an important role in different physiological circumstances including bone remodeling, immune modulation, inflammation and vascularization and pathological conditions such as chronic inflammations, cardiovascular diseases, atherosclerosis, cancer and obesity. OPN has a wide range of biological functions based on its structural modification and the environment in which it is expressed.

**INTRODUCTION**

Osteopontin (OPN) also known as bone sialoprotein I (BSP I), early T-lymphocyte activation-I (ETA-I), urinary stone protein and secreted phosphoprotein 1(SPP 1) is a matricellular phosphoglycoprotein first described in 1979. 1

The word osteopontin is derived from “osteon”, the Greek word for bone and “pons”, the Latin word for bridge indicating its function as a linking protein. 2,3 OPN is produced by a variety of cell types, such as B and T cells, natural killer (NK) cells, macrophages, neutrophils, dendritic cells, bone cells (osteoblasts and osteocytes), breast epithelial cells and neurons. High expression of OPN is detected in the bone, lung, liver, brain, joints, adipose tissue and body fluids such as saliva, blood, urine and milk.4

OPN undergo extensive post translational modification (phosphorylation, glycosylation, sulfation and proteolysis) from different cellular sources, thus OPN has a molecular weight ranging from 41 to 75 kDa, which has a cell type‑specific structure and function. OPN plays a major role in various normal physiological processes, including bone remodelling, immune-regulation, inflammation and vascularisation.5

**STRUCTURE**

OPN gene is located on chromosome 4 region 22 (4q22.1) in humans and contains 7 exons and 6 introns.6 OPN has multiple functional cell adhesive domains including:

1. an arginine-glycine-aspartate (RGD) domain which interacts with cell surface integrins such as αvβ3, αvβ1, αvβ5
2. serine-valine-valine-tyrosine-glutamic acid-leucine-arginine (SVVYGLR) containing domain interacts with α9β1 following exposure by thrombin cleavage
3. calcium binding domain (aa-216-228) and
4. heparin binding domain.7,8

OPN also interact with CD44, the hyaluronic acid receptor.9

OPN has two terminal zones, the N-terminal and C-terminal zones. C-terminal binds two heparin molecules as well as CD44 variants whereas N-terminal includes integrin receptor binding zones.10 OPN is a member of the small integrin-binding ligand N-linked glycoprotein (SIBLING) family. Other 4 members of this family are dentin matrix protein 1 (DMP1), dentin sialophosphoprotein (DSPP), integrin-binding sialoprotein (IBSP), and matrix extracellular phosphoglycoprotein (MEPE).11, 12



**Fig: Structure of OPN showing its functional domains9**

**OPN ISOFORMS**

There are 5 OPN isoforms in humans, formed as a result of alternative splicing of a single SPP1 mRNA transcript 1) full length OPN also known as OPNa - 314aa, 2) OPNb, which lacks exon 5 – 300aa, 3) OPNc, which lacks exon 4- 287aa, 4) OPN4, which lacks exons 4 and 5 – 273aa and 5) OPN5, which contains an extra exon- 327aa.13

**REGULATION**

 OPN expression is affected by a large number of substances including hormones (eg. vit D3 and estrogen) cytokines and growth factors. Inflammatory mediators and growth factors such as interleukin-1 (IL-1), tumor necrosis factor (TNF) and platelet-derived growth factor (PDGF) stimulates OPN transcription via activation of protein kinase C .12 (a) Hormonal and cytokine control of OPN expression-the steroids, retinoic acid and glucocorticosteroid and particularly the seco-steroid hormone vitamin D3 increases OPN expression in bone cells and a marked reduction of OPN mRNA expression is observed in vitamin D3 deficiency. (b) OPN promoter and transcriptional regulation- Increased expression of OPN is associated with an increase in transcription of the OPN gene, which is regulated by transactivation of cis-acting elements in the gene promoter.14

**FUNCTION**

Osteopontin plays important roles in inflammation, biomineralization, wound repair, cardiovascular diseases, cellular survival , cancer, diabetes through different mechanisms.



**Fig: Important biological functions of OPN15**

**OPN IN INFLAMMATION**

OPN is expressed in a variety of immune cells such as macrophages, neutrophils, dendritic cells T and B cells and microglia. It serves as a chemotactic molecule to promote the migration of inflammatory cells under inflammatory condition and acts as an adhesive protein to retain cells at the site. OPN also functions as a pro inflammatory cytokine and can modulate the immune response by enhancing expression of Th1 cytokines and matrix degrading enzymes.16 Plasma OPN level is found to be associated with various inflammatory diseases such as ulcerative colitis and Crohn’s disease.17,18

**OPN IN BIOMINERALIZATION**

 OPN is highly expressed in mineralized tissues such as bone and teeth and is one of the most abundant non collagenous proteins in bone. It is also invariably found in pathological calcifications of soft tissues.7 OPN is found to be expressed by both osteoclast and osteoblasts. Osteoclast derived OPN inhibits hydroxyapatite formation leading to osteoporosis.1

OPN also plays an important role in neuron-mediated and endocrine-regulated bone mass development. The sympathetic nervous system regulates bone mass by changing local bone remodeling through β2-adrenergic receptor . Isoproterenol stimulation of sympathetic nervous system can increase the mRNA and protein levels of OPN in plasma. OPN regulates the ability of β2AR to generate cAMP . Thus OPN participates in the sympathetic nervous system to regulate bone mass through the β2AR/cAMP signaling system. Maintenance of bone homeostasis requires endocrine hormones, including parathyroid hormone (PTH), Klotho, FGF23, and active vitamin D. OPN is an important factor in PTH regulation.19

OPN is also upregulated at sites of pathological calcification such as cardiovascular calcification, urolithiasis.15

**OPN IN CARDIOVASCULAR DISEASES**

OPN is found at the site of atherosclerotic lesions, in association with macrophages and foam cells, indicating that OPN plays an important role in the development and progression of atherosclerosis, vascular remodeling, and restenosis. OPN expression increases after the endothelial lining is damaged by mechanical injury. The process of re-endothelialization of a damaged endothelial lining in an atherosclerotic lesion plays a pivotal role in reducing thrombogenecity. Overexpression of OPN reduced re-endothelialization by inhibiting the migration and proliferation of endothelial cells after injury.20

OPN is also highly expressed in calcified atherosclerotic plaques. It is one of the important negative regulators of calcification, along with matrix glia protein (MGP), fetuin-A and pyrophosphates. OPN directly inhibits calcification as a consequence of tightly binding to hydroxyapatite.21 In contrast to its role of promoting atherosclerotic inflammation, OPN is a potent inhibitor of vascular calcification.22

**OPN IN CANCER**

Initially in the development of tumour cells, local stromal cells express OPN that acts as a signal to attract macrophages and possibly also lymphoctyes. This expression of OPN provides protection against the cytotoxic products of macrophages, while abnormal cells will be killed and removed. However, OPN expression is frequently induced early during the initiation and progression of cancerous growth by transforming agents. High expression of OPN is seen in high-grade metastatic malignant human gliomas.14

Osteopontin mRNA and protein overexpression in several cancers, such as lung23, breast24, prostate cancers25. Osteopontin expression in tumors has been identified by immunohistochemistry, specifically localized in the macrophages in some tumors, and in both tumor cells and macrophages in others.26

**OPN IN DIABETES**

OPN is regarded as a major component in the development of adipose tissue inflammation and insulin resistance and it plays an important role in the pathogenesis of diabetes. Pro-inflammatory cytokines, which play an important role in the development of diabetes complications such as nephropathy, vasculopathy rise in response to OPN release. OPN level also rises in parallel with the severity of diabetes complications.27

OPN is selectively expressed in surrounding inflammatory cells in chronic inflammatory and autoimmune disorders. It is also considered as a secreted sticky molecule that helps in monocyte-macrophage recruitment and control cytokine production in macrophages, dendritic cells, and T-cells. Thus OPN modulation of immune cell response is said to be linked with a variety of inflammatory conditions and may be crucial in the development of adipose tissue inflammation and insulin resistance.28

**OPN IN OBESITY**

Plasma OPN level are significantly higher in overweight and obesity and circulating OPN concentrations correlate with body fat. OPN mRNA and protein are also found to be expressed in omental adipose tissue and this expression is increased in obesity and further elevated in obesity-associated type 2 DM. And it is found that modest diet-induced weight loss is accompanied by a significant decrease in plasma OPN levels.29

Obesity is often found to be associated with non alcoholic fatty liver disease. OPN gene and its receptor CD44 expression in liver were markedly increased and is related to the severity of hepatic steatosis.1

**OPN IN LIVER DISEASES**

In the liver, OPN contributes to the migration of non-parenchymal cells into necrotic areas, and it also serves as an important cytokine - contributing to fibro-genesis. OPN concentration in the plasma was found to predict liver fibrosis in different liver diseases, including: non-alcoholic steatohepatitis (NASH), alcoholic liver disease, as well as in both viral hepatitis B (HBV) and viral hepatitis C (HCV).30,31 Also plasma OPN was found to be elevated in HCC and it has been reported as one of the most promising markers for HCC.32

**OPN IN KIDNEY DISEASES**

OPN mRNA and protein expression is found to be increased in various renal diseases, including stone formation, tubulointerstitial nephritis, glomerulonephritis, acute ischemic renal injury, interstitial inflammation and fibrosis, hydronephrosis, lupus nephritis and many others. And this increase in OPN expression correlates significantly with proteinuria, reduction of creatinine clearance, fibrosis and macrophage and T-cell infiltration.6

**CONCLUSION**

Osteopontin, a multifunctional protein is expressed by various cells of our body. It is involved in normal physiological processes and involved in pathogenesis of variety of disease conditions, including atherosclerosis, cancer, and several chronic inflammatory diseases. OPN is also an important regulator of biomernaralization and potent inhibitor of vascular calcification. OPN increases the inflammation in acute and chronic inflammatory diseases due to its roles in increase of macrophage and T cells in inflammation site.

Osteopontin is an important molecule in field clinical research and can be served as a target for better understanding of the pathophysiology and prognosis of various diseases.

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