**Association of osteopontin promoter polymorphism and aggresivness in breast cancer**

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**Abstract**

Osteopontin (OPN) is an extracellularmatrix protein that is overexpressed in various cancers and promotes oncogenic features including cell proliferation, survival, migration, and angiogenesis, among others. OPN can participate in the regulation of the tumor microenvironment, affecting both cancer and neighboring cells. Here, we review The role of single-nucleotide polymorphism (SNP)-443 of the OPN gene in cancer aggressiveness.

**Introduction**

 Cancer heterogeneity has received increasing attention in recent years. The interindividual [1] and intraindividual [2] variations that are associated with transformation may have implications for prognosis and treatment responses. It is therefore important to understand the consequences of changes in molecules of tumor initiation or tumor progression, such as polymorphisms. The metastasis gene osteopontin (OPN, SPP1)is subject to genetic variation, and variants of the OPN gene — single-nucleotide polymorphisms (SNPs) could contribute to the development and/or progression of specific cancers [3]. In this review, we discuss the association between OPN and cancer, specifically with regard to the roles of SNPs in cancer development, progression and prognosis.

 OPN is a ubiquitous protein produced by a wide range of cell types and tissues, and is found in abundance in body fluids such as blood, milk and urine [4]. The molecule can be presented in soluble form and act as a cytokine, or it can be bound to the extracellular matrix via transglutaminase linkage and promote cell adhesion. OPN plays a key role in tissue remodeling by modulating processes such as bone growth, immune responses, wound healing, cell adhesion, and cell migration. On the molecular level, it interacts with cell surface integrins and CD44, and regulates a wide range of signaling pathways and transcriptional responses [5].When OPN is overexpressed in disease, it contributes to the pathogenesis of atherosclerosis, inflammation, fibrosis, and cancer. Specifically, its association with cancer progression and metastasis has attracted the attention of the scientific community, resulting in the generation of more than 1854 entries for the keywords osteopontin and cancer in PubMed. In general, high levels of OPN correlate with a more aggressive cancer phenotype and are associated with worse prognoses for breast, prostate, liver, head and neck, and lung cancers [6].

**Osteopontin gene SPP1**

OPN is a member of the Small Integrin-Binding LIgand N-linked Glycoprotein (SIBLING) protein family, which is clustered on human chromosome 4, location 4q22.1 The OPN gene is composed of 7 exonsspans 11 [kilobases](https://en.wikipedia.org/wiki/Kilobase%22%20%5Co%20%22Kilobase) in length , 6 of which containing coding sequence encoding the OPN protein with 314 amino acid residues [7].The first two exons contain the 5' untranslated region (5' UTR) [8]. Exons 2, 3, 4, 5, 6, and 7 code for 17, 13, 27, 14, 108 and 134 amino acids, respectively. All intron-exon boundaries are of the phase 0 type, thus alternative exon splicing maintains the reading frame of the OPN gene [9]***.*** Regulation of the osteopontin gene is incompletely understood. Different cell types may differ in their regulatory mechanisms of the OPN gene. OPN expression in bone predominantly occurs by osteoblasts and osteocyctes (bone-forming cells) as well as osteoclasts (bone-resorbing cells) [10] Runx2 (aka Cbfa1) and osterix (Osx) transcription factors are required for the expression of OPN [11] Runx2 and Osx bind promoters of osteoblast-specific genes such as Col1α1,  Bsp, and Opn and upregulate transcription [12]. Hypocalcemia and hypophosphatemia (instances that stimulate kidney proximal tubule cells to produce calcitriol (1α,25-dihydroxyvitamin D3) lead to increases in OPN transcription, translation and secretion [13]. This is due to the presence of a high-specificity vitamin D response element ([VDRE](https://en.wikipedia.org/wiki/VDRE)) in the OPN gene promoter [14]. Extracellular inorganic phosphate (ePi) has also been identified as a modulator of OPN expression [15]. Stimulation of OPN expression also occurs upon exposure of cells to proinflammatory cytokines, [[16]](https://en.wikipedia.org/wiki/Osteopontin#cite_note-pmid11145688-33) classical mediators of acute inflammation (e.g. tumour necrosis factor α [TNFα], infterleukin-1β [IL-1β]), angiotensin II, transforming growth factor β (TGFβ) and parathyroid hormone (PTH), [[17]](https://en.wikipedia.org/wiki/Osteopontin#cite_note-pmid10807582-34) although a detailed mechanistic understanding of these regulatory pathways are not yet known. Hyperglycemia and hypoxia are also known to increase OPN expression [[18].](https://en.wikipedia.org/wiki/Osteopontin#cite_note-pmid10807582-34)

**Osteopontin polymorphism**

 For the human SPP1 gene, the NCBI Single Nucleotide Polymorphism Database (dbSNP) reports 310 sequence variations. Of these 310 variations, 10 correspond to short deletion and insertion polymorphisms and the other 300 are SNPs . 239 of these variations are located in the transcribed region of the gene. Polymorphic sites in the promoter region may impact transcription factor binding and consecutively gene expression. The SNPs for OPN (SPP1) located in the promoter region have been the most studied [19].

 Osteopontin may be induced as a downstream signal transduction target of proto-oncogenic growth factors or secondary to gain-of-function events in transforming signaling pathways . In either case, the binding of cognate transcription factors to spp1 promoter regions is causative for the upregulated expression [20] ***.*** For example, the variant -443 C>T (rs11730582) in the promoter region of OPN gene was found to be located in the transcriptional factor binding site regions, which regulates the transcription of the OPN gene. Recently, the associations between OPN rs11730582 polymorphism and cancer risk have been extensively studied [21] . OPN is a metastasis-related gene that contributes to the progression of over 30 forms of [cancers](https://en.wikipedia.org/wiki/Cancer) , including [lung cancer](https://en.wikipedia.org/wiki/Lung_cancer), [breast cancer](https://en.wikipedia.org/wiki/Breast_cancer) , [colorectal cancer](https://en.wikipedia.org/wiki/Colorectal_cancer) ,  [stomach cancer](https://en.wikipedia.org/wiki/Stomach_cancer), [ovarian cancer](https://en.wikipedia.org/wiki/Ovarian_cancer), papillary thyroid carcinoma,  [melanoma](https://en.wikipedia.org/wiki/Melanoma%22%20%5Co%20%22Melanoma),glioblastoma, osteosarcoma and [pleural](https://en.wikipedia.org/wiki/Pleural)  mesothelioma [20].

### SNPs of human SPP1 gene

 For the human SPP1 gene, the NCBI Single Nucleotide Polymorphism Database (dbSNP) reports 310 sequence variations. Of these 310 variations, 10 correspond to short deletion and insertion polymorphisms and the other 300 are SNPs . 239 of these variations are located in the transcribed region of the gene. 184 correspond to intronic SNPs(one affecting a 3′ end of a splice site), 4 are located in the 5′ UTR and 16 in the 3′UTR, and the remaining 35 in the coding region. From the 71 variations not located in the transcribed region, 58 are in a region approximately 2 kb upstreamof the gene. The remaining 13 variations are in the region 0.5 kb downstream[22] Polymorphic sites in the promoter region may impact transcription factor binding and consecutively gene expression. The SNPs for OPN (SPP1) located in the promoter region have been the most studied [19]***.***

**SNP rs11730582 (−443C<T)**

 rs11730582 is the most studied OPN SNP with the−443 CC genotype generally associated with higher expression of OPN, increased cancer risk ,worse prognosis, and lower survival rate. However in breast cancer and HCC, the−443TT genotype correlates with increased expression of OPN [19]***.*** In breast cancer, the polymorphic site in position−443 of the promoter is associated with tumor grade, such that the allele T is more common in high grade tumors. It is also more common among patients with high OPN levels compared with those with lower OPN levels. The −443 allele T is more common in ER-negative and PR-negative cancers. The importance of the polymorphic site in position−443 may reflect differential transcription factor binding to the distinct alleles. A DNA sequence similar to a c-MYB core binding motif (but not identical to the canonical c-MYB site) immediately precedes the −443 polymorphic promoter position [23]. Transcription via c-MYB from the non-canonical site in the SPP1 promoter may be context-dependent. While c-MYB causes higher transcription from the C allele, there is evidence that under some circumstances the T allele may be associated with higher levels of expression (possibly mediated by a different transcription factor). This implies an important role for c-MYB-independent OPN expression in breast cancer. The differential roles of the SNP in position −443 may be the reason why two meta-analyses of multiple cancer types could not associate this polymorphism with cancer risk [24,25]. In some cancers, MYB may inducehigh level of OPN from theC allele, whereas in other malignancies a yet unidentified transcription factor causes high expression from the T allele [19].

**Conclusion**

OPN rs 11730582 C>T polymorphism is associated with cancer aggressive behavior and poor prognosis in breast cancer patients. Allele T in position -443 is more common and is represented more frequently than the C allele in high grade breast cancer . It is associated with higher aggressiveness of cancer , and consistently hormone receptor- negative cancers tend to grow more rapidly and have a worse prognosis than breast tumors that express ER or PR .

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  **الملخص العربى**

سرطان الثدي هو الشكل الأكثر شيوعا من السرطان في النساء و ثاني سبب رئيسي للوفاة بسبب مرض خبيث في الإناث بعد سرطان الرئة و هو مرض غير متغاير يتضمن العديد من الأنواع الفرعية التي لديها مختلف التكهنات والاستجابات للعلاج.و بسبب الطبيعة السريرية غير المتجانسة لسرطان الثدي، فمن الضروري تحديد المؤشرات الحيوية الجديدة التي ترتبط مع نمو الورم، وتكاثر الأوعية الدموية وانتشار المرض. ونتيجة لذلك، سعت العديد من الدراسات في جميع أنحاء العالم لتحديد أكثر الطرق فعالية لعلاج سرطان الثدي، وتقييم الآثار العلاجية، وتقييم التكهن بشكل صحيح ، و التحديد المبكر للتكرار بعد العملية الجراحية. وقد استخدمت دلالات الاورام الموجوده فى المصل على نطاق واسع كأدوات غير غازية لقياس الاستجابة للعلاج والتشخيص المبكر للتكرار والتنبؤ بحدوث الورم. ويرتبط ارتفاع مستويات osteopontin) ) في أنسجة الورم مع تطور المرض. لذلك ، فإن الكشف عن مستويات osteopontin) ) في مرضى سرطان الثدي قد تكون مفيدة فى التشخيص.و فى نهاية الدرا سه توصلنا الى ان Osteopontin علامة على تقدم سرطان الثدى. ومن المرجح أن المرضى الذين يعانون من مستويات مرتفعة منه فى الدم في وقت التشخيص تستدعي نظم للعلاج أكثر قوة مما هي مناسبة للمرضى الذين يعانون من مستويات منخفضة. كما أنها قد تكون هدف لعلاج سرطان الثدي.