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OPNa variant expression is associated with calcification in thyroid cancer cell line

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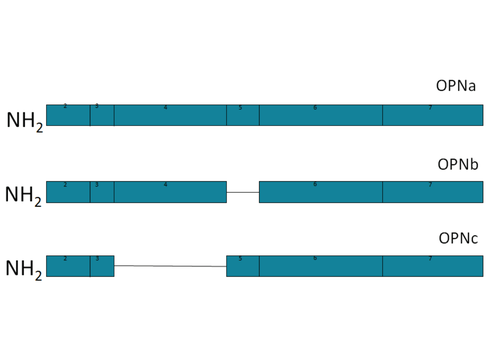
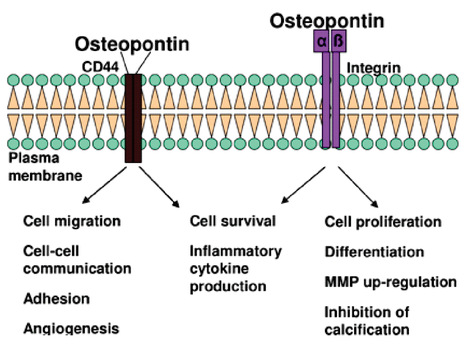
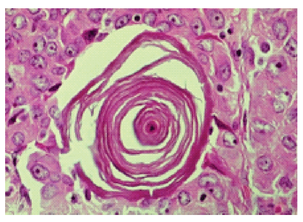
BACKGROUND

Osteopontin (OPN) and its three splice variants (OPN-SV: OPNa, OPNb and OPNc) are overexpressed in several tumors and frequently associated with cancer progression. This holds true for papillary thyroid carcinoma (PTC), which is the most common variant of thyroid cancer (TC), being the histologic type which often presents desmoplasia (collagen deposition) and dystrophic calcification, including a fairly typical feature, the psammoma bodies (PB).





Cancer	Overexpressed isoform	Effects
Prostate	OPNb OPNc	proliferation, migration, invasion
Ovary	OPNc	proliferation, migration, invasion, colony formation
Breast	OPNc	invasion adesion
HCC	OPNa OPNb	migration
Thyoid	OPNa	proliferation, migration, motility, invasion



Psammoma Bodies (PB) – Concentric lamellated calcified structures in PTC

OBJECTIVE

The aim of this study was to investigate the role of OPN-SV expression in the development of PB in classical variant of PTC (cPTC).

METHODS

Total OPN and OPN-SV expression was analyzed by immunohistochemistry and real time PCR in a series of 48 cPTC cases. The association of OPN expression and the presence of PB as well as between PB in cPTC and the clinicopathological features of the tumors were evaluated. TPC-1 and c643 TC cell lines overexpressing OPN-SV were tested for the ability to promote calcification and to synthesize collagen *in vitro.*

RESULTS

Overexpression of OPNa transcripts was significantly associated with the presence of PB in cPTC samples. The presence of PB in cPTC was associated with younger patients and lymph node metastasis. Moreover, OPNa overexpression displayed a strong capacity to promote calcification and substantial collagen synthesis in thyroid cancer cell lines.

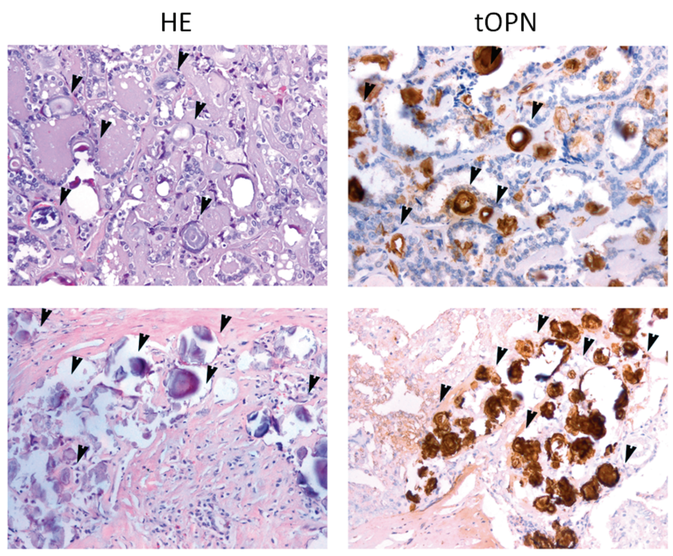


Figure 1. Total OPN staining at PB in cPTC cases. Two different representative cPTC cases showing psammoma bodies (PB) appearing rounded, sometimes fused with each other, or even fragmented, with concentric lamination, as shown by the black arrow heads. PB stained for tOPN antibody, 10 x.

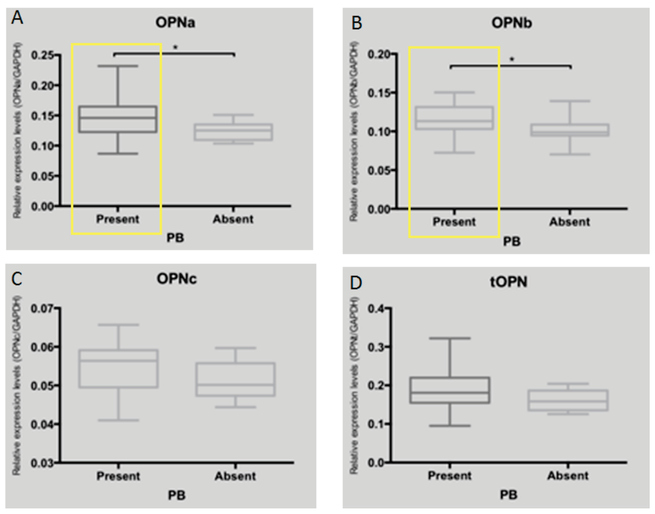


Figure 2. Expression levels of OPNa, OPNb, OPNc and tOPN transcripts in cPTC concerning presence or absence of PB. **(A)** OPNa **(B)** OPNb **(C)** OPNc and **(D)** tOPN mRNA expression levels measured by real time PCR * $p < 0.05$. Results are from at least two independent assays with triplicates.

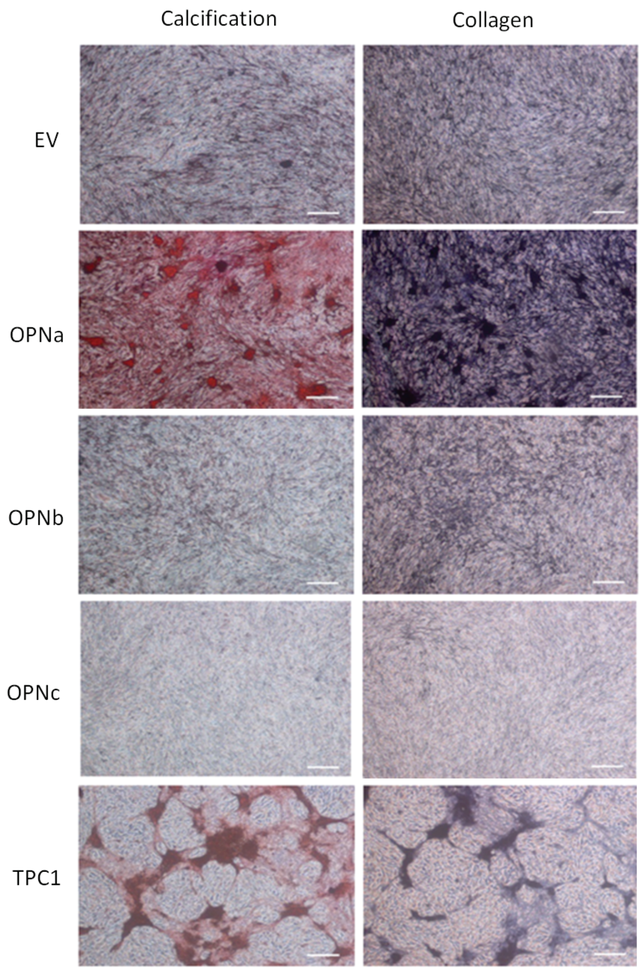


Figure 3. Calcification and collagen production in c643 cells overexpressing OPNa, OPNb, OPNc, EV and TPC1 cells. Left panel: Matrix calcification detected with Alizarin Red staining. Dark orange areas correspond to extracellular matrix (ECM) rich in calcium deposits. Right panel: Collagen ECM production was determined by Masson trichrome staining. Dark purple areas correspond to ECM rich in collagen. Scale bar: 100 µM. Representative photomicrographs of 2 independent experiments at 24 days of culture are shown.

CONCLUSIONS

Our data suggest that OPNa plays a role in the formation of PB often associated with cPTC. Basic research on the interactions between OPNa overexpression by tumor cells and the surrounding microenvironment may give clues for a better understanding of cPTC biology and phenotype.

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