**DNA methylation in the upstream CpG island of the GPER locus and its relationship with ERα expression in breast cancer.**

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**Abstract**

The seven-transmembrane G-protein coupled estrogen receptor (GPER) relays short-term non-genomic responses in target cells and tissues. Clinical investigations have indicated lower GPER expression in breast tumors than in normal tissues, and its expression is negatively associated with tumorigenesis. Based on these findings, GPER is predicted as a potential tumor suppressor. Hypermethylation of the promoter region is a known mechanism for silencing tumor suppressors. GPER expression significantly correlates with a number of clinicopathological markers, such as ERα and PR. Given either negative or positive association between GPER and ERα expression in breast cancer, the current investigation examines the association between upCpGi methylation of GPER locus and ERα expression in breast cancer. Using the targeted bisulfite sequencing approach, we have reported that DNA methylation in the upCpGi of the GPER locus is, at least in part, responsible for the loss of GPER expression in ERα-negative breast cancer cell lines or breast tumors. We also described a differentially methylated region (DMR) comprising of terminal eight CpG dinucleotides in the upCpGi, where most methylation was observed. The likely association between methylation and ERα expression in clinical breast tumor samples may serve as an indirect indicator of GPER expression in tumors. Unlike ER, PR and HER2, the detection of GPER by immunohistochemistry is difficult due to the absence of high-quality standardized antibodies. Hence, PCR or sequencing-based methods to estimate upCpGi methylation may be used to predict the GPER expression in tumor biopsies.

**Keywords**: GPER, upCpGi, Bisulfite sequencing, DMR