3OST3B與CDH3基因於台灣乳癌檢體組織之甲基化變異及基因功能分析

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ABSTRACT

The death rate of breast cancer in Taiwan has obviously risen these years, which becomes a very important issue. Many risk factors such as aberrant DNA methylation of promoter region of tumor suppress gene and growth-related gene have been reported, besides family history of breast cancer, endocrine, and carcinogen. Therefore, this study was focused on the relation between aberrant DNA methylation in gene promoter region and breast cancer tumorigenesis. Tumor and normal tissues obtained from 76 breast cancer patients were used to analyze the methylation status of promoter region and gene expression in both of cell membrane protein gene－P-cadherin (CDH3) and regulatory extracellular matrix glycoprotein gene－heparan sulfate D-glucosaminyl 3-O-sulfotransferase (3OST3B) by methylation specific PCR(MS-PCR) and combined bisulfite restriction analysis (COBRA). Because there was no aberrant methylation observed in CDH3, only 3OST3B was chosen for further analyses including COBRA- sequencing, reverse transcription PCR (RT-PCR), and immunohistochemical stain (IHC). The distribution of methylated CpG site and the expression of mRNA and protein were analyzed. According to the methylation status of 3OST3B in the tumor tissues, we divided the 76 patients into three groups－ 27 patients of hypomethylation, 20 patients of intermedium-methylation and 29 patients of hypermethylation. In 3OST3B promoter region, there were many CpG dinucleotides found in the binding sites of several transcription factors such as NF-κB, E2F, and n-MYC. The methylation status of CpG dinucleotides might affect the expression of 3OST3B. The results of IHC showed that the expression of 3OST3B in tumor tissues with 3OST3B hypermethylation were lower than in normal tissues. After the treatment of demethylation agent, 5-azadC, in breast cancer cell line MDA-MB-231, the methylation level of 3OST3B decreased and the gene expression was restored. The association of 3OST3B methylation status with clinicopathological features of patients with breast cancer showed that 3OST3B with intermedium-methylation and hypermethylation was significantly correlated with non-metastasis (M0) and high p53 expression. Therefore, 3OST3B could be a valuable candidate gene as a predictor of breast cancer risk assessment.

Keywords : CDH3 ; 3OST3B ; DNA Methylation ; COBRA ; breast cancer

Table of Contents

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