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Title: Profile of WT1 methylation in human lung cancer

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Body: Background: CpG island hypermethylation of the promoters of the genes is a well-known mechanism of genetic alteration involved in carcinogenesis. Wilms tumor gene (WT1) is involved in the regulation of human cell growth and differentiation and it is a modulator of oncogenic K Ras signaling in lung cancer; methylation of WT1 was never studied in human lung cancer. The primary aim of this study is to compare the methylation profile of WT1 promoter in samples of neoplastic and non-neoplastic lung tissue taken from the same patient. Methods: Lung tissue obtained from 16 patients with non small cell lung cancer (NSCLC) and 6 patients with small cell lung cancer (SCLC). The methylation status of 29 CpG islands in the 5' region of WT1 by means of pyrosequencing was investigated. Results: The mean percentage of methylation, considering all CpG islands of WT1 in the neoplastic tissues of all NSCLC patients, was 16.2 ± 3.4 , whereas in the control tissue from the same patients it was 5.6 ± 1.7 ($p < 0.001$), adenocarcinomas present greater methylation vs squamous cell carcinomas ($p < 0,001$). In SCLC was not statistically significant difference between neoplastic tissue and control tissue of same patients. Conclusions: Although, WT1 methylation does not seem to play a role in the pathogenesis of SCLC, hypermethylation of WT1 seems to be involved in the carcinogenesis of NSCLC. CHT, histotype, exposure to cigarette smoke are all factors influencing the gene CpG islands which become hypermethylated in NSCLC.