Effect of 5-aza-2'-deoxycytidine on Estrogen Receptor Alpha/Beta and DNA Methyltransferase 1 Genes Expression, Apoptosis Induction, and Cell Growth Prevention of the Colon Cancer HT 29 Cell Line

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Abstract

Background: Cellular activity such as gene expression is regulated by epigenetic mechanisms and modifications. In mammals, DNA methylation is an essential component of the epigenetic machinery of the cells. DNA hypermethylation of the several tumor suppressor genes (TSGs) is associated with transcriptional gene silencing resulting in colon tumorigenesis. Overexpression of DNA methyltransferase 1 (DNMT1) in colon cancer has been reported in several studies. The methylation of estrogen receptor alpha (ERα) and estrogen receptor beta (ERβ) have been demonstrated in various cancers. Previously, we indicated that genistein can reactivate ERα in hepatocellular carcinoma (HCC). The present study was designed to investigate the effect of 5-aza-2'-deoxycytidine (5-aza-CdR) on ERα/ERβ and DNMT1 gene expression, apoptosis induction, and cell viability inhibition of the colon carcinoma HT 29 cell line.

Methods: The effect of 5-Aza-CdR on the colon carcinoma HT 29 cell viability was measured by MTT assay. To determine the apoptotic cells, the cells were assessed using the Annexin V-FITC/PI detection kit. The expression of ERα, ERβ, and DNMT1 genes was determined using real-time quantitative RT-PCR.

Results: The results indicated that 5-Aza-CdR can inhibit cell growth significantly versus control groups, induce significant apoptosis, down-regulate DNMT1, and up-regulate ERα and ERβ genes expression at different time periods. The percentage of apoptotic cells was 85.83% and 86.84% after 24 and 48 h, respectively ($P < 0.01$). The IC50 value for 5-Aza-CdR was obtained at 2.5 μM.

Conclusions: 5-Aza-CdR can up-regulate ERα and ERβ genes expression through DNMT1 down-regulation resulting in apoptosis induction and cell growth prevention.

Keywords: 5-aza-2′-deoxycytidine; colon cancer; epigenetic.