Investigation of the Effect of Zebularine in Comparison to and in Combination with Trichostatin A on p21Cip1/Waf1/ Sdi1, p27Kip1, p57Kip2, DNA Methyltransferases and Histone Deacetylases in Colon Cancer LS 180 Cell Line

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Abstract

Background: The heart of the cell cycle regulatory machine is a group of enzymes named cyclin-dependent kinases (Cdks). The active form of these enzymes includes a kinase and its partner, a cyclin. The regulation of cyclin-Cdk complexes is provided by Cdk inhibitors (CKIs) such as Cip/Kip family comprising p21Cip1/Waf1/Sdi1, p27Kip1, and p57Kip2. The hypermethylation and deacetylation of Cip/Kip gene family seem to be frequent in numerous cancers. It has been indicated that increased expression of DNMTs and HDACs contributes to cancer induction. Previously, we reported the effect of DNA demethylating agents and histone deacetylase inhibitors on histone deacetylase 1, DNA methyltransferase 1, and CIP/KIP family in colon cancer. The current study was designed to evaluate the effect of zebularine in comparison to and in combination with trichostatin A (TSA) on p21Cip1/Waf1/Sdi1, p27Kip1, p57Kip2, DNA methyltransferases (DNMT1, 3a and 3b) and histone deacetylases (HDAC1, 2, and 3) genes expression, cell growth inhibition and apoptosis induction in colon cancer LS 180 cell line.

Materials and methods: The colon cancer LS 180 cell line was cultured and treated with zebularine and TSA. To determine cell viability, apoptosis, and the relative expression level of the genes, MTT assay, cell apoptosis assay, and qRT-PCR were done respectively.

Results: Both compounds significantly inhibited cell growth, and induced apoptosis. Furthermore, both compounds increased p21Cip1/Waf1/Sdi1, p27Kip1, and p57Kip2 significantly. Additionally, zebularine and TSA decreased DNMTs and HDACs gene expression respectively.

Conclusion: The zebularine and TSA can reactivate the CIP/KIP family through inhibition of DNMTs and HDACs genes activity.
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Keywords: Colon cancer; TSGs; Zebularine; trichostatin.