PP-296

Investigation of the Anti-Tumourogenesis Potentialof 5-Aminosalicylic Acid: Lack of Efficacy in Caco-2 Cells

ElifKALE[1], Fatma SANDAL[1], Alaattin ŞEN[1]
[1]Department of Biology/Faculty of Arts & Sciences, Pamukkale University, Turkey
fatmasandal@windowslive.com

Aim of the study: Recent trials and epidemiological researchesrecommend that nonsteroidal anti-inflammatory drugs are effective in the counteractive action of various malignancies. Inflammatory bowel disease and ulcerative colitis patients are likely to have an expanded hazard for the development of intestinal tumours. 5-Aminosalicylic (5-ASA) is ananti-inflammatory drug regularly utilized as a part of the treatment in these patients and may give reliance against the improvement of colorectal malignancy in these patients. For this purpose, this study was designed to investigate the anti-tumorigenesis activitiesof 5-ASA on theexpression level of some tumorigenesis related genesin Caco-2 cells.

Material and Methods: The human epithelial colorectal adenocarcinoma cells line (Caco-2) was purchased American Tissue Culture Collection.Caco-2cellswere grown in monolayer culture in DMEM medium containing 10% FBS, 1% penicillin/streptomycin at 37°C in a humidified atmosphere comprised of 95% air and 5% CO_2. Cell viability was assessed using WST. Caco-2 cellswere treated with varying concentrations of 5-ASA at mM range for 24 h at 37 °C.Total RNA was isolated using RNaesy Mini Kit (Qiagen) by the manufacturer's standard protocol.Quantitative Real Time PCR (qRT-PCR) analysis was performed using KiloGreen qPCR Master Mix in an Exicycler 96 Real Time Quantitative Thermal Block PCR System to determine p53, Rb-1, Cyclin D1,Cyclin D2, CDKN1A, PTEN, Myc and Jun gene mRNA expression level.

Results: Two different concentrations of 5-ASA (20 and 50 mM)were identified to measure the differential responses of Caco-2 cells. p53 and PTEN genes mRNA expression levelwere decreased 2.19, 2.47 and 2.41, 3.56 times in Caco-2 cellsas a result of 20 and 50 mM concentrations of 5-ASA treatments, respectively.Similarly, Rb-1 gene mRNA expression level was decreased 1.03 and 1.22 times, respectively. Myc genes mRNA expression levels were decrease 1.67 and 1.45 in Caco-2 cells with20, 50 mM doses of 5-ASA,respectively. Cyclin D1 and Cyclin D2 genes mRNA expression levels were lowered 1.26, 2.69 and 4.5, 4.35 in Caco-2 cells as a result of 20 and 50 mM concentrations of 5-ASA treatments, respectively. CDKN1A gene mRNA expression level was decreased 1.08 and 1.43 times in Caco-2 cells with 20 and 50 mM 5-ASA applications, respectively. Jun gene mRNA expression level was decreased 1.11 and 1.17 times in Caco-2 cells with 20 and 50 mM 5-ASA applications, respectively. To summarize,mRNA expression levels of all genes were downregulated. Based on these results, we suggest that 5-ASA does not have any anti-tumorigenesis effect in Caco-2 cells and does not give any protections against the colorectal malignancy in these patients.

Acknowledgments: This work supported by Pamukkale University PAUBAP-2015FBE042

Keywords: 5-aminosalicylic acid (5-ASA),Caco-2,Anti-tumorigenesis,