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Anticancer effect of postbiotic derived from fermented milk of *Lactobacillus helveticus* MTCC 5463 on HT-29

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There is increasing interest in postbiotics as potential therapeutic agents due to their biofunctional properties. As non-viable bacterial products or metabolites that offer health benefits, their anticancer potential warrants investigation. This study focuses on examining the effects of postbiotics derived from lactic acid bacteria fermented milk, specifically from *Lactobacillus helveticus* MTCC 5463, on colon cancer cells. This study screened postbiotics (cell-free supernatants) derived from the fermented milk of four LAB strains for various biofunctional activities. The most promising postbiotic, derived from *Lactobacillus helveticus* MTCC 5463 was then assessed for its anticancer effects on the HT-29 colon cancer cell line using the MTT assay. The study used the RT-PCR method to examine the impact of the postbiotic on the expression levels of genes associated with apoptosis, including *Bax, Bcl-2, Caspase-8, cyclin D1*, and *p53*. After 24, 48, and 72 h of treatment, the IC50 values of postbiotic were 3.0, 1.5, and 1.0 mg/mL, respectively. Gene expression analysis via RT-PCR revealed upregulation of pro-apoptotic genes (*Bax* and *Caspase-8*) and downregulation of antiapoptotic genes (*Bcl-2. cyclin D1*, and *p53*) in HT-29 cells treated with the postbiotic compared to untreated control cells. These findings suggest that the postbiotic derived from the fermented milk of probiotic MTCC 5463 could serve as a promising biological agent against colon cancer.

Keywords: Cell-free supernatant, Colon cancer, Lactic acid bacteria, Gene expression, Apoptotic