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Cytotoxic Effect of Biologically Transformed Propolis on HCT-116 Human Colon Cancer Cells

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Abstract : Object: Propolis which consists of compounds that are accepted as antioxidant, antimicrobial, antiseptic, antibacterial, anti-inflammatory, anti-mutagenic, immune-modulator and cytotoxic, is frequently used in current therapeutic applications. However, some of them result in allergic side effects, causing consumption to be restricted. Previously our group has succeeded in producing a new biotechnological product which was less allergenic. In this study, we purpose to optimize production conditions of this biologically-transformed propolis and determine the cytotoxic effects of obtained new products on colon cancer cell line (HCT-116). Method: Firstly, solid propolis samples were dissolved in water after weighing, grinding and sizing (sieve-35mesh) and applied 40 kHz/10 min ultrasonication. Samples were prepared according to inoculation with Lactobacillus plantarum in two different proportions (2.5% and 3.5%). Chromatographic analyzes of propolis were performed by UPLC-MS/MS (Waters, Milford, MA) system. Results were analysed by UPLC-MS/MS system MassLynx™ 4.1 software. HCT-116 cells were treated with propolis examples at 25-1000 µg/ml concentrations and cytotoxicity were measured by using WST-8 assay at 24, 48, and 72 hours. Samples with biological transformation were compared with the non-transformed control group samples. Our experiment groups were formed as follows: untreated (group 1), propolis dissolved in water ultrasonicated at 40 kHz/10 min (group 2), propolis dissolved in water ultrasonicated at 40 kHz/10 min and inoculated 2.5% L. plantarum L1 strain (group 3), propolis dissolved in water ultrasonicated at 40 kHz/10 min and inoculated 3.5% L. plantarum L3 strain (group 4). Obtained data were calculated with Graphpad Software V5 and analyzed by two-way ANOVA test followed by Bonferroni test. Result: As a result of our study, the cytotoxic effect of propolis samples on HCT-116 cells was evaluated. There was a 7.21 fold increase in group 3 compared to group 2 in the concentration of 1000 µg/ml, and it was a 6.66 fold increase in group 3 compared to group 1 at the end of 24 hours. At the end of 48 hours, in the concentration of 500 µg/ml, it was determined 4.7 fold increase in group 4 compared to group 3. At the same time, in the concentration of 750 µg/ml it was determined 2.01 fold increase in group 4 compared to group 3 and in the same concentration, it was determined 3.1 fold increase in group 4 compared to group 2. Also, at the 72 hours, in the concentration of 750 µg/ml, it was determined 2.42 fold increase in group 3 according to group 2 and in the same time, in the concentration of 1000 µg/ml, it was determined 2.13 fold increase in group 4 according to group 2. According to cytotoxicity results, the group which were ultrasonicated at 40 kHz/10min and inoculated 3.5% L. plantarum L3-strain had a higher cytotoxic effect. Conclusion: It is known that bioavailability of propolis is halved in six months. The data obtained from our results indicated that biologically-transformed propolis had more cytotoxic effect than non-transformed group on colon cancer cells. Consequently, we suggested that L. plantarum-transformation provides both reduction of allergenicity and extension of bioavailability period by enhancing healthful polyphenols.

Keywords : bio-transformation, propolis, colon cancer, cytotoxicity
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