

Testing the Activity of Epigallocatechin-3-gallate on Cells of Colorectal Carcinoma in vitro

Glezi, Viliam (School: Gymnazium Ludovita Stura)

The research was aimed at testing the effects of the polyphenolic compound Epigallocatechin-3-gallate on three colorectal cancer cell lines. The effects of this substance have been observed in the cosmetics industry, and several studies have been published which are discussing the effect of the substance on urogenital and breast cancers. All cell lines examined were derived from primary colon adenocarcinoma, with the HT-29 / EGFP / FUR line being specific for its resistance to 5-Fluorouracil. Cells were cultured with various concentrations of EGCG in triplicate in a 96 well plate and observed by real-time microscopy in an InCuCyte ZOOM Imaging System. After 72 hours, we evaluated confluence and found that some concentrations of EGCG significantly inhibited tumor cell growth, including the chemoresistant line. In contrast, low concentrations had the opposite effect and even promoted proliferation of these cells. A concentration of 50 $\mu\text{g} / \text{ml}$ was considered toxic because of complete growth inhibition. Necrotic cells were observed during microscopic examination. Based on the results, we tried to prove the mechanism of action of EGCG on tumor cells in terms of molecular biology. Using quantitative RT-PCR, we examined the effect of EGCG on the inactivation of transmembrane transport proteins from the ATP binding cassette family, where we found that as the concentration of EGCG increased, the expression of genes encoding the proteins decreased. It is believed that reducing the expression of these proteins, which transport the drug into the extracellular space, reduces the chemoresistance of these cells. We also performed a viability assay by cytometry and as another prove we incubate chemoresistant tumor cells with EGCG and chemotherapeutic 5-FU.