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Summary of doctoral thesis of **MICHAŁ MATYSIEWICZ**

„*ROLE OF SELECTED OPIOID PEPTIDES AND CYTOKINES IN HUMAN COLORECTAL CANCER*"

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Each year, about one million new cases of colorectal cancer are diagnosed in the world. According to the recent findings, all over the world, is diagnosed about 6% new cases of the disease – 40% of these patients die. Colorectal cancer is the third most common malignant cancers among men and the second most among women. The exact causes of colon cancer are not known. However, the main risk factors include external (environmental influences and eating habits) and internal (other diseases and genetic factors) factors. It is believed that opioid peptides may play a role in the etiology of colon cancer. Among them, beta-endorphin is the most frequently mentioned one. It is believed that it affects nervous system resulting in the formation of acetylcholine and norepinephrine. The neurotransmitters affects the immune system by activating immune cells to direclyt fight the cancer or to indirect secretion of cytokines. Among the most frequently mentioned cytokines are: pro-inflammatory interleukin-6 (IL-6), tumor necrosis factor α (TNF-α) and interleukin-1β (IL-1β) and anti-inflammatory interleukin-10 (IL-10).

The aim of the study was to determine the concentration of beta-endorphin and selected cytokines in serum, urine and tumor tissues obtained from 110 patients with colorectal cancer and to determine changes in the functioning of the tumor cells of colorectal cancer models: Caco-2, HT-29 cells and healthy colon epithelial: CCD 841 CoTr, under the influence of acetylcholine, norepinephrine and beta-endorphin.

Studies were carried out in three stages. First (1) the levels of beta-endorphin and selected cytokines (IL-6, TNF-α, IL-1β and IL-10) in clinical material collected from patients diagnosed with colorectal cancer was determined. Secondly (2) the effect of neurotransmitters

and beta-endorphin on featured colon cell lines in vitro was studied. The changes in the secretion of cytokines, cell proliferation (BrdU assay), cell viability (NR uptake assay) and changes in MOR gene expression was also investigated. In the third section (3) effect of acetylcholine, norepinephrine and beta-endorphin on the immune system (PBMC) of patients with colorectal cancer was determined in vitro. Furthermore, the dual cell in vitro model which demonstrated the influence of neurotransmitters and beta-endorphin on the proliferation of Caco-2, HT-29 and CCD 841 CoTr cells, through the immune system cells (PBMC) was designed.

Studies revealed the presence of beta-endorphin both in the tissues obtained from patients with colorectal cancer and in the healthy controls. IL-6 levels in serum, urine and tissues of cancer patients were elevated in proportion to the stage of the cancer. Neurotransmitters and beta-endorphin significantly influenced the changes in the proliferation of tumor cells, but did not alter proliferation of healthy epithelium of the colon. Acetylcholine, norepinephrine and beta-endorphin affected viability of tumor cells. It was also shown that the tested substances had cytotoxic effect on healthy cells CCD 841 CoTr. The test substances affected the changes in MOR gene expression. MOR expression decreased in Caco-2 cells under the influence of acetylcholine and increased under the influence of norepinephrine and beta-endorphin. In the case of HT-29 cells, MOR gene expression was reduced after stimulation with all the tested substances. However, the CCD 841 CoTr cells showed an increase in MOR expression under the influence of norepinephrine. The test substances (neurotransmitters and beta-endorphin) caused increase of IL-6 secretion by PBMC isolated from patients with colorectal cancer. In the dual cell in vitro model, it has been demonstrated that stimulation of immune cells (PBMC), by acetylcholine, norepinephrine, and beta-endorphin significant decreases proliferation of Caco-2 and HT-29 cells. These changes were not observed in the case of healthy cells.

Ever increasing problem of colorectal cancer compel scientists to search for new diagnostic and pharmacological solutions. This study will hopefully contribute contribute to widening the knowledge about the biochemical processes taking place in the body of patients with colorectal cancer.