Mgr Maciej Piotr Frant
Zakład Wirusologii i Immunologii,
Instytut Mikrobiologii i Biotechnologii,
Wydział Biologii i Biotechnologii,
Uniwersytet Marii Curie-Skłodowskiej w Lublinie

Lublin, 09.03.2016

Ocena aktywności wybranych porfiryn w stosunku do ludzkich komórek raka jelita grubego w badaniach *in vitro*

(Assessment the activity of selected porphyrins in human colorectal cancer cells *in vitro*)

Abstract

Medical conditions associated with a large intestine, mainly cancers, represent a very serious social and economic problem. Yearly about one million of new cases of a colorectal cancer is being diagnosed worldwide. This disease is located on a third place in terms of incidence in men (660 000 cases/year), and on a second in women (570 000 cases/year) around the world. According to estimates of the International Agency for Research on Cancer in Europe (EU), the colorectal cancer is the most common cancer, which is the second most common cause of death. There are many reasons for the development of the colorectal cancer. Factors, such as improper diet, lack of physical activity, obesity, occupational factors, genetic load, alcohol consumption, smoking, pollution, infections, tendency to constipation or age, have a negative effect on the structure and function of the large intestine leading to the development of cancer. In addition, research suggests that inflammatory mediators present in the microenvironment may favor the development of the colorectal cancer.

In the treatment of the colorectal cancer both primary and secondary prevention are very important. The development of disease is preceded by a small dysplastic change in the intestinal epithelium. Subsequently, a gradual increase of such a change occurs resulting is adenoma, which in time acquires invasive characteristic. The detection of cancer at its earliest stage of development (secondary prevention) increases the chances of a patient's survival. In case of the colorectal cancer three standard therapies are used: surgery, chemotherapy and radiotherapy,

used alone or in combination. Conventional therapies of the colon cancer do not give a full guarantee of a recovery. Currently in Poland, the curability of the colorectal cancer is only 30-35%, while in the foreign facilities (United States of America and Western Europe) it reaches 65%. Therefore, the introduction of the new methods of treatment and increasing public awareness about the risks of the colorectal cancer, in combination with the developed prevention, may be significant and most importantly - effective impulse contributing to improve such statistics.

Among the alternative methods of cancers treatment, such as the colorectal cancer, the photodynamic therapy (PDT) is often mentioned and indicates a significant efficacy in case of certain types of cancer. It uses an interaction of three different factors: light, photosensitizer and oxygen. Light induces a photosensitizer, which transmits the excitation energy to oxygen, generating the formation of reactive oxygen derivatives. The photodynamic therapy often uses porphyrins - natural organic compounds with a tetrapyrrole structure. Due to tendency to accumulate in the cells, simplicity of excitation by the light of a suitable wavelength and the ability to generate reactive oxygen species, porphyrins seem to be a very good photosensitizer in the fight against cancer. There is a significant potential for the use of this particular group of compounds in photodynamic therapy, not only in case of superficial tumors but also those less accessible, like the colorectal cancer.

The aim of this study was to determine whether the selected porphyrin derivatives, after exposure to white light, have significant anti-tumor activity in a cellular model of the colorectal cancer. For this purpose, an attempt to assess the cytotoxic activity of the test compounds to CCD 841 CoTr cells (normal epithelial cells of the large intestine) and HT29 cell line (colorectal cancer), to determine the porphyrin localization in these cells and the effect of porphyrin to cell migration and a tumor microenvironment was made.

A strong toxicity of the manganese porphyrin in tumor cells and weaker toxicity in normal cells was proven. In addition, there were no changes in cytotoxicity after light induction in the analyzed model. A reverse situation occurred in case of the free base porphyrin, which showed negligible cytotoxicity in the absence of light exposure, however after light induction the cytotoxicity was increasing. The cell localization of free base porphyrin was determined. Initially, it was outside the area of the cytoplasm and later, around the nucleus. The influence of porphyrins to inhibition of cell migration (mainly normal cells) was proven. Analysis

of inflammatory factors (IL-1β, IL-6, IL-10, IL-6Rα, COX-2) revealed a wide range of effects that porphyrin have to cell metabolism and tumor microenvironment. Magnesium porphyrin did not show photodynamic nature, but due to its high cytotoxicity, high for the malignant cells, could be used as an anticancer agent. Free base porphyrin, due to its photodynamic activity could be a useful photosensitizer in the potential treatment of early stages of the colorectal cancer. However, further studies to determine the efficacy of the free base porphyrin *in vivo* model are needed.

In conclusion, porphyrin derivatives seem to be an interesting group of compounds for the further study. The obtained results have confirmed the anti-cancer potential against colorectal cancer cells, which give base to further studies to determine molecular mechanisms of action of this group of compounds.