LA.1

ORP150 prevents collagen degradation in glucose-deprived breast cancer MCF7 cells

Marzanna Cechowska-Pasko¹, Rafał Krętowski¹, Edward Bańkowski²

¹Department of Pharmaceutical Biochemistry, ²Department of Medical Biochemistry, Medical University of Białystok, Białystok, Poland

e-mail: Marzanna Cechowska-Pasko <mapasko@gmail.com>

In normal mammary gland tissue basement membrane (BM) is a continuous layer that separates epithelial cells from the surrounding stroma. Collagens are the main components of BM. They are the first proteins that must be degraded in order to start the neoplastic cell invasion. It has been described that type IV collagen plays an important role in cell adhesion and migration, ERK tyrosine phosphorylation and activation of matrix metalloproteinases. Glucose is the main energetic substrate for tumour cells. We decided to study the effect of glucose deprivation on collagen synthesis and degradation in breast cancer MCF7 cells, and a correlation of these processes with the expression of oxygen/glucose regulated proteins (ORP150/GRP170). The incorporation of radiolabeled proline into collagenase-sensitive and hydroxyproline-containing proteins was used as an index of collagen synthesis, whereas pulse-chase technique was employed to evaluate the degradation of newly synthesised proteins. We demonstrated that MCF7 cells incubated in high glucose medium synthesised detectable amounts of collagenous proteins. Most of them were found in the cell layer. The shortage of glucose resulted in about 30% reduction in synthesis of collagenous proteins, both those secreted into culture medium and remaining in the cell layer. The pulse-chase experiments demonstrated that the reduced amount of newly synthesised collagen was protected against intracellular degradation. Proportionally less collagen was degraded in cultures incubated in low glucose than in high glucose medium. These phenomena were accompanied by an increase in the expression of endoplasmic reticulum chaperon - ORP150 in cultures growing in low glucose medium. We suggest that the increased expression of ORP150 is a factor which protects collagen against intracellular degradation induced by glucose deprivation.

LA.2

The effect of polyphenols - tannic acid, resveratrol and its derivatives on the induction of apoptosis in rat C6 glioma cells

Małgorzata Zielińska-Przyjemsk¹, Violetta Krajk-Kuźniak¹, Agnes Rimando², Wanda Baer-Dubowska¹

¹Department of Pharmaceutical Biochemistry, Poznan University of Medical Sciences, Święcickiego 4, 60-781 Poznań, Poland; ²Natural Products Utilization Research Unit, Agricultural Research Service, U.S. Department of Agriculture, University, USA;

e-mail: Małgorzata Zielińska-Przyjemsk <mzielin@ump.edu.pl>

Gliomas are the most common malignant primary brain tumors characterized by widespread invasion throughout the brain, destruction of normal brain tissue and resistance to traditional therapy. Thus development of new agents able to reactivate cell death programs, particularly apoptosis, is important for introducing new therapeutic and/or chemopreventive strategies for malignant gliomas. Our earlier studies showed that tannic acid and naturally occurring stilbenes: resveratrol and its analogue pterostilbene, the common ingredients of berries fruits affected the key events involved in the carcinogenesis process including induction of apoptosis in human polymorphonuclear leukocytes. The aim of this study was to evaluate the effect of these compounds on rat C6 glioma cell line proliferation and their ability to induce apoptosis. As showed the results of MTT assay all polyphenols including resveratrol synthetic derivative - 3',5,4'-trimetoxytiblene (TMS) were cytotoxic at the concentrations range above 50 μM. The increase of phosphatidylserine externalization and loss of mitochondrial membrane potential detected by flow cytometry, the measures of apoptosis, were observed after 24 hours of treatment with tested compounds at the concentrations of 1 and 10 μM. Western blot analysis showed increased p53 level after 48h of treatment. Tannic acid and TMS were the most efficient apoptosis inducers exerting the effect comparable to camptothecin used as reference compound. These results indicate that naturally occurring polyphenols and their synthetic derivatives, particularly tannic acid and TMS may be considered as potential chemotherapeutic and/or chemopreventive agents for treatment malignant gliomas.
The influence of a diet supplementation with methionine on the morphological picture of the lungs and pancreases of rats intoxicated with sodium fluoride

Barbara Stawiarska-Pięta1, Anna Marek1, Ewa Grucka-Mamczar2, Ewa Birkner2, Jolanta Zalejska-Fiolka2
1Department of Pathology, Faculty of Pharmacy in Sosnowiec, Silesian Medical University in Katowice, Poland; 2Department of General Biochemistry, Faculty of Medicine in Zabrze, Silesian Medical University in Katowice, Poland
e-mail: Barbara Stawiarska-Pięta <bspieta@o2.pl>

In the previous studies, the pathomorphological changes appear in many organs, including the lungs, liver, kidneys and heart of rats intoxicated with fluoride. Studies in recent years showed that in the pathomechanism of observed pathological changes, a disturbance of oxidoreductive processes played a crucial role. Past experiments have shown that the modifications of antioxidant defense system (enzymatic and nonenzymatic) in fluorosis were observed. The simultaneous administration of vitamin E and C in animals intoxicated with fluorine increases the activity of antioxidative enzymes and lowers MDA levels. The aim of the study was to examine the influence of methionine on the development of pathomorphological changes in the morphological picture of rats’ organs intoxicated with sodium fluoride.

These results indicate that naturally occurring polyphenols and their synthetic derivatives, particularly tannic acid and TMS may be considered as potential chemotherapeutic and/or chemopreventive agents for treatment malignant gliomas.

The study was performed on Wistar 18 rats (adult female). The animals were divided into 3 groups: control group - which were kept on distilled drinking water, studied group I - where animals were given sodium fluoride solution to drink in doses of 10.0 mg NaF/kg body mass/24h, and studied group II - in which animals were given sodium fluoride solution to drink in doses of 10.0 mg NaF/kg body mass/24h, being on a diet supplemented with methionine in the amount of 2mg/rat/24h. The experiment lasted 35 days. Upon dissection, the lungs and pancreases were taken for histopathological examination, and the blood was collected for biochemical analyses. The pathomorphological changes in the organs were assessed on the basis of preparations obtained by the normal paraffin method, stained with hematoxylin and eosin (H-E). Protein concentration in the pancreases was determined by means of Lowry's method, whereas the aldolase activity (ALD) in the blood serum was determined by colorimetric method.

In the case of NaF administration, pathomorphological examinations of lungs revealed that examined rats developed erythorrhagia, hyperemia, inflammatory infiltrations and emphysematous blebs. There was a focal vacuolar degeneration of cells and inflammatory infiltrations noticed in pancreases. The results show the positive but not fully effective influence of methionine on the changes caused by fluoride in rats’ organisms. Methionine prevented the fluoride interference in glycolysis but did not have any effect on the protein concentration caused by fluoride. The results confirmed the antioxidative properties of methionine.