P4

Comparative analysis of adaptor protein CIN85/Ruk isoforms expression in human benign prostate hyperplasia and prostate adenocarcinoma

O.I. Basaraba[1]*, Ya.P. Bobak[2], G.Yu. Shuvayeva[2], V.L. Buchman[3], L.B. Drobot[1]. [1]*Palladin Institute of Biochemistry of the National, Laboratory of Cell Signalling, Kyiv, Ukraine,* [2]*Institute of Cell Biology, Deparnnent of Cell Signaling, Lviv, Ukraine,* [3]*Cardiff University, Neuroscience Group, School of Biosciences, Wales, UK*

Adaptor proteins play an important role in facilitating protein-protein interactions and subsequent formation of signalling networks. These proteins recruit binding partners to a specific location inside the cell, and also regulate their activity. Adaptor protein Ruk/CIN85 is important components of different regulatory pathways involved in control of cell proliferation, adhesion, invasion and survival, and, thus, an play a role in uterine carcinogenesis. Neoplastic transformation of prostate tissue includes a broad set of oncological diseases, but the molecular mechanisms of these pathological processes remain unclear.

The main goal of our research was to study cin85/ruk gene expression in samples of human benign prostate hyperplasia (BPH) and prostate adenocarcinoma both at the level of mRNA and protein using Northern blot and Western blot analysis.

Using Northern blot analysis, one cin85/ruk mRNA transcript of approximately 3.2 kb was detected in analyzed samples of prostate tumors. This mRNA transcript codes for full-length form of adapter protein CIN85/Ruk with molecular weight of 85 kDa. Both BPH and adenocarcinoma samples were characterized by polymorphism in the expression level of 3.2 kb mRNA transcript of cin85/ruk. Using anti-RukS Western-blot analysis, multiple molecular forms of CIN85/Ruk with molecular weights of 140, 130, 100, 85 and 50 kDa were detected in the samples of BPH. Pattern of multiple molecular forms of CIN85/Ruk in prostate adenocarcinoma samples differed from the previous ones by appearance of the additional immunoreactive bands corresponding to p70, p56, p40 and p34. Our data suggest that the pattern of Ruk/CIN85 expression depends on specific molecular features characteristic for individual samples.

The obtained results suggest that changes in the expression level of cin85/ruk mRNA transcripts and multiple molecular forms of CIN85/Ruk in BPH and adenocarcinoma samples can lead to the loss of coordinated control of apoptosis and proliferation in the transformed cells. These data will offer new opportunities for the identification and validation of key molecular tumor targets to be exploited for novel therapeutic approaches.

P5

Is obesity changing the expression profile of genes coding IGF in the colorectal cancer patients?

M. Muc-Wierzgon[1]*, E. Nowakowska-Zajdel[1], R. Braczkowski[2], T. Kokot[1], U. Mazurek[3], M. Rudzki[4], D. Waniczek[4]. [1]*Medical University of Silesia, Department of Internal Medicine, Bytom, Poland,* [2]*Medical University of Silesia, Department of Public Health, Bytom, Poland,* [3]*Medical University of Silesia, Department of Molecular Biology, Sosnowiec, Poland,* [4]*Medical University of Silesia, Department of Surgery, Bytom, Poland*

Epidemiological researches indicate that obesity is a risk factor of colorectal cancer. The fatty tissue is a place of synthesis and excretion of many cytokines as IGF1, TNFα, IL-6, VEGF, TGFβ, leptin, adiponectin and others.

The aim of the study was to analyse mRNA expression profile of genes coding IGF in relation to body mass index (BMI) in the colorectal cancer patients.

Material and Methods: The colon cancer specimens were taken during surgery treatment of 35 colorectal cancer patients (22 men; 13 women, aged 65.5$\pm$8.9). Examined patients were divided into I–IV groups according to TNM Classification (I-7, II-9, III-10, IV-9 patients). They were divided into A and B groups as well, in relation to BMI (A: BMI $<$ 25) – 17 patients, B: BMI $\geqslant$ 25) – 18 patients). A number of mRNA copies of genes coding IGF1, IGF2, IGF1R and IGF2R were examined with QRT-PCR method. The experiments were performed according to the protocol approved by the Ethic Committee of the Medical University of Silesia in Katowice.

Results: There were no statistical differences of mRNA copies of genes coding IGF1, IGF1R, IGF2, IGF2R in the tumor tissue between examined groups A and B. But the analysis showed that the number copies of mRNA IGF1 was enough higher in the patients with overweight and obesity than in the group with normal weight (15052$\pm$4077 versus 8558$\pm$2409). The level of mRNA genes coding IGF were similar in tissues representing different clinical staging but the analysis showed the higher number copies of IGF1, IGF1R, IGF2 according to advancement of cancer. There were no correlation between the number of mRNA copies of genes coding IGF and BMI in the group A. But there was negative correlation between the number of IGF1 and BMI ($R = 0.6727$; $p = 0.330$) and positive correlation between the number of IGF2R and BMI ($R = 0.5441$; $p = 0.0238$) in the group B.

Conclusions:

1. The changes of expression profile of genes IGF and its receptors in colorectal cancer patients according to body mass were found.
2. The analysis should be done among patients according to level of advancement of cancer disease, taking into consideration body mass index for the same clinical staging.
3. The findings suggest that the changes of expression profile of genes coding IGF could be connected with autocrine and paracrine function of tumor cells in colorectal cancer.

Acknowledgements: This study was supported in part by a Ministry of Scientific Research and Information Technology Grant NN404167234.

P6

"AminoIndex" for cancer detection (2): plasma free amino acid profiling for breast cancer screening

H. Yamamoto[1]*, N. Okamoto[2], A. Chiba[3], Y. Miyagi[2], A. Imaizumi[4], T. Ando[4], M. Onuma[1], M. Yamakado[5], A. Yoshida[3]. [1]*Ajinomoto Co., Inc., HI (Health Informatics) Department, Tokyo, Japan,* [2]*Kanagawa Cancer Center, Cancer Prevention and Cancer Control Division, Yokohama, Japan,* [3]*Kanagawa Cancer Center, Department of Breast and Thyroid Surgery, Yokohama, Japan,* [4]*Ajinomoto Co., Inc., Institute of Life Sciences, Kawasaki, Japan,* [5]*Mitsui Memorial Hospital, Center for Multiphasic Health Testing and Services Tokyo, Japan*

Introduction: Amino acids balance is changed in patients of various diseases due to metabolic transition while it is maintained in healthy human such as various cancers. We previously demonstrated that significant changes of plasma amino acid profile was observed and classifier composed of plasma amino acid concentrations as explanatory variables ("AminoIndex") showed high discrimination ability for breast cancer patients (Okamoto 2009). In this study, further possibilities of "AminoIndex" for breast cancer were investigated.

Subjects and Methods: Venous blood samples were collected from Japanese breast cancer patients before any medical treatment (N = 109). Those of controls were also collected from subjects who were undergone comprehensive medical examination at Mitsui Memorial Hospital (N = 1,699). After plasma separation, amino acid concentrations were measured by LC-MS.

60 patients and 300 age-matched control subjects were chosen as study data set to predict "AminoIndex". And