activities, total glutathione, concentration of TBA-reactive substance (TBARS), protein carbonyls and oxyradical formation. Metallothioneins concentration as a quantity of thiol groups (MT-SH) was evaluated. Molecular markers of toxicity [DNA strand breaks, cholinesterase activity, lactate dehydrogenase] were assessed. The set of biomarkers of stress and toxicity was applied due to the guidelines. The low density (LDL) and high density (HDL) lipoproteins, glycated hemoglobin (HbA1c), total cholesterol and triglycerides were also measured.

In obese patients, the concentration of total cholesterol (by 29%) and lower density lipoproteins (LDL, by 59%) is higher, while the high-density lipoproteins (HDL, by 19%) is lower than in the control group. The ratio of HDL/LDL in the control group is 0.82 and decreases for obesity to 0.44. Parameters of lipid metabolism correlate with BMI (r [0.73], p<0.001). The concentration of triglycerides and glycosedylated hemoglobin is similar in both groups of patients.

The lower activity of superoxide dismutase, the higher activity of catalase and the level of oxyradicals formation in the obese patients (O-group) were shown compared to the control group which persons had no appropriate pathology (C-group). The investigated O-group was characterized by the lower concentration of glutathione and the higher concentration of metallothioneins. In obese patients, oxidative stress (integral index of oxidative stress = - 0.43), as well as signs of geno- and neurotoxicity were manifested by increasing the DNA fragmentations and cholinesterase activity respectively. The correlation between the concentration of metallothioneins and oxidative stress indices in the regression model was existed: MT-SH = 0.027×OR + 0.41×CAT− 0.55×SOD*, R² = 0.92; F(3,12)=57.8, p<0.001.

Results. The use of the principal component analysis with the NIPALS algorithm allowed to find correlations between investigated parameters of the examined normal-weight and obese individuals. Metallothioneins form a joint cluster with parameters of oxidative stress, cytotoxicity, index of body mass, total cholesterol, and low density lipoprotein. These indices are also crucial in the development of obesity, as they correlate with O-group with a high significance. The control group is located in opposition to the O-group and includes indices of reduced glutathione and high density lipoprotein. This arrangement proves the relation of the patterns within the cluster and their opposite nature between the two clusters. To sum up, the integrated analysis of the selected parameters of lipids’ metabolism and oxidative stress, metallothioneins and signs of cytotoxicity in obese young women allowed to determine the amount of features that deepens pathological changes. It is the discrepancy in oxidative-reductive status related to simultaneously activation of catalase and manifestation of oxidative injury, the redox-equilibrium shift in the direction of the prooxidant processes and the disturbance of the balance of anaerobic / aerobic glycolysis and NAD+/NADH, reducing the portion of high-density lipoproteins to low-density lipoproteins, as well as increasing the DNA fragmentation. Metallothioneins have a partial tread effect on radical processes and reduce manifestations of oxidative damage to biomolecules in obese patients. Comparatively the same capacity of metallothioneins we have shown before for human thyroid nodular goiter and ovarian cancer model (Falfushynska et al., 2014, 2015). In this way metallothioneins should be deeply studied as a putative supplier for obese patients in a reason of oxidative injury consequences relieve.

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References


Key words: obesity, oxidative stress, lipoproteins, metallothioneins, cytotoxicity

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VITAMIN D: A POTENTIAL BIOMARKER FOR MORE AGGRESSIVE PROSTATE CANCER

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One of the many functions of vitamin D is associated with its antiproliferative and proapoptotic effects on prostate cancer cells. One of the debatable questions nowadays is whether vitamin D deficiency can lead to an increased risk of prostate cancer. Recent studies have shown that low serum vitamin D levels may be associated with worse prognosis in cancer patients. It could be assumed that vitamin D may serve as an important biomarker of prostate cancer aggressiveness. However, levels of vitamin D are not routinely measured in daily clinical practice. In Bulgaria more complex and multi-faceted studies on vitamin D status and its relationships with clinical determinants in prostate cancer have not been conducted. The aim of the present study was to perform a comparative analysis of vitamin D status in prostate cancer patients with different aggressiveness and to assess the relationship with clinical and biochemical parameters characterizing the disease.

Materials and Methods. This prospective study included 88 men with histologically proven prostate cancer (52 - 85 years) attending the Urology Clinic at University Hospital - Varna in the period January-December 2015. The
tumor aggressiveness, tumor invasion and metastatic potential, and risk for biochemical recurrence were evaluated according the EUA guideline, 2016. PSA was determined immunochemically, serum 25-hydroxy vitamin D (25OHD) levels were assayed by HPLC-MS/MS (Galunska et al, 2014), bioavailable 25OHD was calculated according Kim et al, 2017. VDBP and albumin were analyzed using commercial kits. One-way ANOVA, correlation and multifactorial regression analysis were used for data analysis. Statistical significance was accepted at p<0.05.

**Results.** Worsened vitamin D status demonstrated by significantly lower levels of total 25OHD and bioavailable 25OHD was established for prostate cancer patients, compared to the control noncancerous patients (p < 0.001 for 25OHD, p < 0.05). A decreased total and bioavailable 25OHD with the risk of bone metastases (PSA>20ng/ml) was indicated. The risk of biochemical recurrence negatively correlates with the serum total (Spearmn r = - 21, p =0.05) and bioavailable 25OHD (Spearmn r = - 34, p =0.01). Serum 25OHD was lower in patients with more aggressive prostate cancer and reversibly correlated with the Gleason score (Spearmn r = - 0.26, p <0.05). Multiple regression analysis showed negative influence of serum 25OHD on tumor aggressiveness (B = - 0.24, p<0.05). ROC-curve analysis revealed better diagnostic efficiency for 25OHD (AUC = 0.69, p<0.001) compared to the widely accepted as a biomarker total testosterone (AUC = 0.65, p<0.01).

Further research for validation and introducing serum 25OHD levels to other biomarkers already established and used in clinical practice as well as changing the algorithm of treatment by vitamin D supplementation of patients with prostate diseases.

**References:**

**Key words:** prostate cancer, vitamin D, tumor aggressiveness, biomarker

**RELATIVE GENE EXPRESSION OF SELECTED MIRNAS AS POSSIBLE BIOMARKERS IN COLORECTAL CANCER STAGE DEVELOPMENT**

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Many studies have demonstrated the significant contribution of miRNAs in cancer pathogenesis - controlling tumor growth, invasiveness and avoidance of the immune system by cancer cells.

**Materials and Methods.** In this preliminary study, we observed miR-143, miR-200c, miR-101, miR-34b and miR-375 gene expression in 42 samples of formalin fixed and paraffin embedded colorectal cancer (CRC) tissue samples in comparison to healthy neighboring tissue of same patients. Expression levels were analyzed in relation to RAS mutational status and cancer stage. Gene expression analyses were performed using qPCR technique and expression levels were normalized against U6.

**Results.** The results of RAS wild type CRC patients showed decreased expression of all investigated miRNAs in contrast to RAS mutant patients, where no change in selected genes was detected. According to cancer progression stage, change in the expression profile of miRNAs was observed. In stage IIA levels of expression of all studied miRNAs were reduced compared to healthy tissue controls. MiR-200c and miR-101 were elevated more than twice approximately, whereas miR-143, miR-34b and miR-375 were not changed in stage IIIB CRC. At stage IV, which is related to tumor metastasis decreased levels of for miR200c, miR101 and miR375 were established and no change in the profile of miR143 and miR34b expression. The fact that miR143 changes only in stage IIA patients is possibly due to accumulation of mutations in RAS genes in late stages (IIIB and IV). The observed increase in miR-200c expression in IIIB stage may be related to increased metastatic potential in nearby lymph nodes and organs. MiR-101 has the same expression profile as miR-200c in different stages of CRC. The decrease of miR-375 levels in early stage IIA possibly aims to support tumor proliferation.

Among the analyzed in recent study miRNAs, miR-200c and miR-101 appeared to be the most promising candidates for potential biomarkers, for stage development of colorectal cancer, in future analyses.

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**Keywords:** colorectal cancer, miRNAs, gene expression

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