УДК 615.356:616–006

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ВИТАМИНЫ, КАРОТИНОИДЫ И МИКРОЭЛЕМЕНТЫ В ДИНАМИКЕ ОНКОГЕНЕЗА ПРЕДСТАТЕЛЬНОЙ ЖЕЛЕЗЫ

VITAMINS, CAROTENOIDS AND MICROELEMENTS IN PROSTATE CARCINOGENESIS

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Статья поступила в редакцию: 13.01.2015
Статья принята к печати: 19.02.2015

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The article received: 13.01.2015
The article approved for publication: 19.02.2015

Аннотация. Проблема эпидемиологии и профилактики рака предстательной железы (РПЖ) в течение многих десятилетий остаётся актуальной и нерешённой. Один из аспектов этой проблемы – роль различных пищевых добавок и диеты, различающихся для людей разных регионов. На фоне многочисленных и неоднозначных эпидемиологических и клинических исследований по данной проблеме необходимы дальнейшие клинико-экспериментальные работы, посвящённые изучению роли витаминов, витаминоподобных, биологически активных веществ и микроэлементов на разных этапах инициации и формирования рака простаты. Использован комплекс клинических, гистологических методов обследования урологического больного и методы анализа в крови витаминов и родственных им соединений, а также продуктов перекисного окисления липидов и 21 микроэлемента. Объекты исследования – 115 мужчин-пациентов клиники урологии Первого Московского государственного медицинского университета им. И.М. Сеченова, из них 95 пациентов с различной онкопатологией простаты и 20 мужчин без выявленных заболеваний этой железы. Доказаны статистически достоверные следующие изменения концентрации исследованных веществ в крови. У больных с РПЖ уменьшено содержание антиоксидантов – ликопина, витамина Е, германия, селена и увеличено содержание прооксидантов – алюминия и диеновых конъюгатов. Для пациентов с простатической интраэпителиальной неоплазией высокой степени (ПИН ВС) доказано уменьшение в крови содержания витамина С, суммарных каротиноидов, германия и селена и увеличение – алюминия и диеновых конъюгатов. При раковом перерождении клеток простаты уровень в крови антиоксидантов (ликопина, каротиноидов, витаминов Е и С, германия, серы, селена) понижен, а для прооксидантов (алюминия и диеновых конъюгатов) повышен. Показанные изменения, связанные с механизмом канцерогенеза железы, зафиксированы уже стадии предрака (ПИН ВС) без дальнейшего их усиления на стадии сформированного рака простаты.

Annotation. Epidemiology and prophylaxis of prostate cancer (CaP) has remained a pressing and challenging issue for many decades. One of its aspects is the role of food additives and diet which differ for people of various regions. Multiple dubious epidemiologic and clinical studies on this issue warrant further clinical and experimental research into the role of vitamins and microelements in different stages of prostate cancer initiation and development. The use of complex clinical and histological evaluation of urologic patients and blood sampling is viewed for vitamins as well as lipid peroxidation products and 21 microelements. The test subjects included 115 male patients of the Urology clinic – 95 patients with different kinds of prostate gland oncopathology and 20 males without prostate pathology. The results have shown a statistically significant deviation of the levels of certain substances in the blood. Patients with CaP had reduced levels of antioxidants (lycopene, vitamin E, germanium, selenium) and increased levels of pro-oxidants (aluminum and conjugated dienes). We have demonstrated that patients with high-grade prostatic intraepithelial neoplasia (HGPIN) have decreased blood levels of vitamin C, total carotenoids, germanium, selenium and increased levels of aluminum and conjugated dienes. Malignant transformation of prostate gland cells is associated with a decreased blood level of antioxidants (lycopene, carotenoids, vitamin E, vitamin C, germanium, sulfur, selenium) and a increased level of pro-oxidants (aluminum and conjugated dienes). The aforementioned changes associated with the mechanism of prostate carcinogenesis are already detected in patients with HGPIN (precancer) with no further progression in patients with developed CaP.

Ключевые слова. Рак предстательной железы, предрак, витамины, каротиноиды и микроэлементы крови

Keywords. Prostate cancer, precancer, vitamins, microelements.

INTRODUCTION

The preventive and therapeutic effect of vitamins, vitamin-like substances, and microelements on prostate cancer (CaP) development has long been a debatable issue. There is a sufficient amount of experimental, epidemiologic, and clinical evidence showing anticancer (mainly preventive) properties of vitamins E (tocopherols), C, D, A (retinol), carotenoids (β-carotene, lycopene) and other substances and their effects on CaP. This data correlates to the influence of dietary habits in various pats of the world on CaP incidence. Thus, a diet rich in different antioxidants (vitamins E, C, A, carotenoids, selenium) and phytoestrogens (isoflavones) that is characteristic of Asian and Mediterranean peoples correlates to a lower CaP incidence than that in Western Europe and the USA [1-3]. Several works and even reviews do not support these findings [4, 5, 6], and if they do they are limited to certain conditions. For instance, vitamin E in smokers or β-carotene in males with low dietary intake of carotene may decrease the risk of CaP development [7].

Several recently published clinico-epidemiological trials of large populations with a follow-up of several (up to 12) years provided even more contradictory conclusions about the effects of various dietary supplements (vitamin E, vitamin C, selenium) on CaP development [6, 8-10]. For instance, a daily allowance of 400 IU of vitamin E (for a follow-up of a minimum of 7 and maximum of 12 years) was found to lead to a 17% increase in prostate cancer incidence among healthy men as compared to the placebo group [9]. Another report of the same (SELECT) trial also showed that vitamin E intake increased the risk of CaP among men with lower baseline selenium status, but had no effect among men with high selenium status [8]. Such an effect produced by vitamin E (tocopherol with three unsaturated bonds) can be explained by its participation in the lipid peroxydation (LPO) of membrane phospholipids, where this vitamin acts as a pro-oxydant, not as an antioxidant. Surprisingly, the authors of the above-mentioned paper found evidence that vitamin E increased the risk of CaP among men with high selenium status, and came to a conclusion

that men should avoid perennial vitamin E and selenium supplementation at doses exceeding recommended dietary intakes. As a matter of fact, biochemists have been studying the interaction between vitamin E and selenium (present in selenocysteine of glutathione peroxidase), as well as their synergistic and dissimilar effects, for a long time. Vitamin E supplementation is known to reduce the need for selenium present in glutathione peroxidase, and in this respect the explanation provided in [8] for the roles of vitamin E and selenium in CaP prevention at the level of LPO appears to be valid.

Some researchers put forward recommendations on the vitamin D supplementation doses in the narrow range associated with cancer prevention, and reported that both high and low vitamin D concentrations were associated with the risk of CaP [10]. The data reported in [10] appear to support our earlier findings: on the level of transcription vitamin D and its derivatives (being steroids) may have uncertain effects on the regulation of proliferation and apoptosis in prostate epithelium [11].

Over the last years (2011-2013) closer attention has been paid to CaP prevention during precancer formation – high-grade prostatic intraepithelial neoplasia (HGPIN) – and its transformation into CaP [12-15]. Various rates of progression of HGPIN into CaP have been reported – 36% within a 3 year-long monitoring period in one case [16]. It has been suggested to employ chemoprevention of CaP, use therapeutic strategies to delay and stop the transformation of HGPIN into CaP or even facilitate regression of HGPIN during these stages of carcinogenesis [16-17]. The use of special selenium compounds (selenomethionine, methylseleninic acid), vitamin E, vitamin D, lycopene, soy isoflavones, polyphenols contained in green tea (some of which are antioxidants), and statins – cholesterol synsthesis inhibitors – has been predicted to be effective [18-20].

However published experimental, clinical, and epidemiologic data concerning this issue also remain dubious thus far. It may be due to the fact that the extent of influence of vitamins, microelements, and other factors on CaP initiation, formation, and development depends on multiple hereditary and other factors which are not usually taken into account when conducting studies of the aforementioned design. The effects of vitamins and their analogues taken in physiological doses are not limited to antioxidant properties. They are able to activate transcription factors and cause other epigenetic changes [4, 21]. In general terms it can be narrowed down to the widely accepted fact of general biochemistry that vitamins and metals act as components of coenzymes and cofactors which in turn enter the composition of many enzymes – complex proteins. Apart from that, in the majority of the above-mentioned epidemiologic and clinical studies the administration of additives – vitamins, minerals, and other substances – is not followed by monitoring of their concentrations in the body (in the blood, biopsy samples or tissue samples obtained during surgery). Some studies include blood sampling for assessment of serum levels of vitamins and vitamin-like substances which allows gaining insight into anticarcinogenic effects of these antioxidants [10, 22-25].

One more drawback of the analyzed works should be noted. They mainly state the incidence of CaP and its change with food additive use and diet, lacking the analysis of vitamin and metal concentration in the blood as mentioned above. In such cases the study is limited to formed CaP, usually localized CaP; it is seldom that research focuses on the influence of vitamins on untreated CaP progression [5]. We did not find any published papers in with to study changes in the content in the body (blood) of vitamins, vitamin-like substances, and microelements on prostate cancer initiation and progression. However, as we carried out our project at the Sechenov First Moscow State Medical University, we had at our disposal all the state-of-the-art laboratory facilities and equipment one would need to conduct a model study of prostate carcinogenesis and analyze clinical, urological, biochemical, and histological data obtained from healthy test subjects and patients with benign prostatic hyperplasia (BPH), low-grade prostatic intraepithelial neoplasia (LGPIN), HGPIN (i.e. precancer), and CaP.

The research project was divided into several cycles and focused on vitamins, vitamin-like substances, and microelements with conditional simulation of prostate carcinogenesis and evaluation of the aforementioned substance levels in the blood. Some of the results were published earlier as short reports [26-31].

MATERIALS AND METHODS

Before their blood levels of vitamins and microelements were measured, participants underwent a clinical and laboratory examination that involved prostate biopsy. Following that, they were divided on the basis of the results obtained (primarily, on the basis of the histological analysis of the prostate biopsy specimens) into groups depending on the marked morphological changes in patients, i.e. signs of CaP, or HGPIN (precancer), or LGPIN. Most patients also had other (secondary) types of morphological changes in the prostate that are mentioned in the tables below. For the second research cycle a control group was formed.

The described division of patients into groups according to their prostate biopsy results is further substantiated by our findings that show varying blood levels of certain substances in these groups: starting with group 4 (control group) and moving on to group 3 (LGPIN), then to group 2 (HGPIN), and finally to group 1 (CaP), we observed higher levels of PSA and aluminum, yet lower levels of lycopene and germanium (see tables describing the second research cycle). Lycopene levels decrease more than twice, whereas aluminum levels increase 10 to 15 times. We believe that these biochemical data indirectly confirm sufficient methodological level our histology laboratory.

The first research cycle was conducted during winter and spring on 21 patients with prostate pathology and did not include a control group of healthy test subjects. The normal concentration ranges of serum levels of vitamins and carotenoids in healthy individuals recommended by the Scientific Research Institute of Nutrition were considered normal concentration ranges for this study [32].

The second more thorough research cycle was conducted one year later during winter, spring, and summer on 94 males, residents of the Moscow region. The test subjects were divided into 4 groups following their clinical and laboratory examination. All the patients underwent simultaneous evaluation of microelements levels in the blood.

Patient groups:

1. 1st group (26 patients) – CaP as the main clinical and histological diagnosis in some cases associated with PIN, BPH, and chronic prostatitis (CP).

2. 2nd group (26 patients) diagnosed with HGPIN (precancer) in some cases associated with LGPIN, BPH, and CP.

3. 3rd group (22 patients) – LGPIN, BPH, CP or their combination based on histological examination. This group may be considered as a "symbolical control group" for the previous two.

All these males (aged 35 to 81 years, mean – 64 years) underwent biopsy or transurethral resection of the prostate (TURP) followed by histological examination of the biopsy specimens.

4. 4th (control) group consisted of 20 younger males (aged 25 to 63 years, mean – 43 years) admitted to the Urology clinic for examination or treatment of other urological pathologies (urolithiasis, hydronephrosis, chronic pyelonephritis). No significant prostate pathology was detected in these patients. The group included healthy volunteers as well as official Urology clinic patients.

Exclusion criteria were tumors of other organs and additional intake of other multivitamins, 5α-reductase inhibitors, male sex hormones or their analogues, antiandrogens, and gonadotropin-releasing hormone agonists (gonadoliberin agonists).

All the patients underwent standard complex diagnostic evaluation including complete blood count, biochemical assay, total PSA level assessment, urinalysis, three-glass test, uroflowmetry, digital rectal examination (DRE), and transrectal ultrasound imaging of the prostate (TRUS) followed by PSA density (PSAD) calculation. In some cases an additional examination – dynamic magnetic resonance prostatovesiculography (DMRP) – was performed. Depending on the results of prostate cancer screening (PSA level, PSAD, as well as DRE, TRUS, and DMRP results) the patients underwent transrectal ultrasound-guided multiple (8 to 12 sites) needle prostate biopsies from standard sites and also sites where CaP foci were suspected on TRUS (hypoechoic areas), and DMRP. Biopsies were taken using a "B&K 3535" (BK-Medical) diagnostic ultrasound system with a 6558/T/S bi-plane sector mechanical rectal probe operating at a frequency of 7.5 MHz. Apart from biopsy samples, tissues obtained during TURP were also used.

The histological examination of prostate biopsy specimens and tissues obtained during TURP included standard tissue processing (paraffin wax embedding), 4 μm section preparation followed by hematoxylin and eosin, pickrofuchsin staining. In complicated cases an immunohistochemical analysis using Dako antibodies to prostate-specific antigen (PSA) and to Dako p63 protein was performed. p63, which is a p53 tumor suppressor homolog, is being constantly secreted by the basal layer cells of the prostate epithelium, and it is frequently used in difficult cases to distinguish between CaP and PIN [14]. Universal criteria and the Gleason grading system were used for CaP evaluation. Criteria for differentiation between CaP, PIN, BPH and CP were also used [33-35]. We consider it necessary to emphasize that precise differentiation between HGPIN and LGPIN was conducted in accordance with histological criteria developed by Bostwick DG, Brawer MK (1987) modified by Joniau S et al. 2006 [14] or other researchers [35].

The concentrations of the studied substances (vitamins, carotenoids, lipid peroxidation products) were assessed (using a standard protocol) by a group of experienced researchers working at the Scientific Research Institute of Nutrition, who co-authored this article. The blood was drawn in the medical facility into blood collection tubes containing separator gel without filler (additives). The test tubes were then centrifuged for 10 min at 1500 RPM. The obtained serum was cooled and delivered to the laboratory within one hour. Vitamin C concentration was visually identified in fresh serum with the help of visual titration with Tillman's reagent [32]. The serum for assessment of vitamin A, vitamin E, and carotenoids was stored at -20°C for no longer than one month. Lipid peroxidation products – conjugated dienes– were detected spectrophotometrically. Malonic dialdehyde was detected with the help of thiobarbiturate assay [32]. Concentrations of fat-soluble vitamin A (retinol), vitamin E (tocopherols), and carotenoids were determined using high-performance liquid chromatography (HPLC) according to the described method [36] with some changes.

Briefly, 200 μL of serum was treated immediately after obtaining with antioxidant butylated hydroxytoluene (BHT) and 200 μL of methanol was added; the mixture was shaken in a vortex mixer for 30 s and 200 μL of *n*-hexane was added. The tube was shaken for 20 min and centrifuged at 3000 rpm for 10 min. 120 μL of the hexane extract was evaporated under nitrogen to remove the solvent. The residue was redissolved in 20 μL of dichloromethane and 100 μL of acetonitrile-methanol (1:1, v/v) was added. 50 μL of this sample solution was analyzed with HPLC, using column (150 x 4.6 mm, i.d.) with Nucleosil 100-5 C18 (Macherey-NAGEL

GmbH & Co. KG, Germany); 880-PU pump (Jasco, Japan); eluent: acetonitrile-methanol-dichloromethane (50:45:5, v/v/v), at a flow-rate of 0,7 mL·min^{-1}. We used two sequentially connected detectors (Jasco, Japan): a 870-UV spectrophotometer at 450 nm for determination of the carotenoids and an 821-FP fluorescence spectrophotometer with a programmable variable wavelength : initial λ_{ex} 325 nm, λ_{em} 480 nm – for the assay of retinol; after 5 min of the run the λ_{ex} was changed to 295 nm and λ_{em} was changed to 330 nm for γ- and α-tocopherols. The recoveries were: retinol – 93%, and tocopherols – 96 %, lycopene – 104 %, β-carotene – 102%. The measurements of fat-soluble vitamins and carotenoids were performed in triplicate. All reagents were purchased from Sigma Chemical Co.

The analysis of chemical elements was performed by one of the authors of this article (Barashkov G.K.), who has a vast experience in the field [37]. Freshly drawn venous blood (1 mL) for assessment of 21 chemical elements was put into a plastic test tube with 200 units of Na-heparin and stored at -20°C. Na-heparin level was taken into account during further calculation. Once the necessary amount of samples (no less than 10) was accumulated, the material underwent analysis [37]. The samples were transferred into teflon crucibles containing a mixture of 1 mL of concentrated nitric acid ("high purity") and 3 mL of 37% hydrogen peroxide. The mixture was burned in steam table (marmite) for 2 h. After that 5 mL of 5% nitric acid was added to the breakdown product which was analyzed. Microelements were analyzed using ICP-OES 'Optima-3000' atomic emission spectrometer produced by the PerkinElmer Company. When this type of spectrometer is used, blank readings (used for detection of possible traces of metals in vials and reagents) are being automatically subtracted from sample readings. Thus, such traces (if present) could not invalidate the sample results. When preparing vials and choosing reagents, we followed the guidelines listed in the spectrometer manual.

The statistical analysis was performed according to the statistical guidelines for biological studies accepted in Russia and published by the Russian Academy of Sciences, the Russian Academy of Medical Sciences, and the Belarusian "High School" [38-41]. ([40] was co-authored by one of the authors of the present article.) All of the mentioned Russian guidelines agree well with those developed in other countries and do not contradict them.

The main reason why we used statistics in our research was to find out whether there were any differences between each of the 30 blood levels of substances studied in the four groups of urological patients. Given a large amount of the available data, we did not intend to use statistical techniques for anything else. In our work, we used Student's t-test (comparison of the mean values of two independent samples) as the main method to identify statistically-significant differences between the arithmetic mean values, which characterize the blood concentrations of one specific substance between patients of each of the four groups. This test can be used when dealing with both large and small sample sizes (usually more than 20 variants), and when the number of variants is different in different groups [38,39,41]. According to I.P. Ashmarin, A.A. Vorobyov [39], and V.Yu. Urbach [38], when the number of variants in each group exceeds 20, this test "may be used to carry out final statistical processing" [39, p. 32]. With 20 variants in each group, the number of the degrees of freedom will be f=20+20-2=38, which corresponds to the critical values of t=2,02 (when the probability of error is p<0,05), and t=2,70 (when p<0,01). The critical values of t are shown in the tables included into some guidelines on statistical analysis |38, 39, 41|. One can compare the application of Student's t-test to this pair of mean values with the critical values of t listed in the tables, so as to be able to make a conclusion.

This test has been widely used for decades in scientific researches of the Russia and in research projects implemented across the world.

RESULTS AND DISCUSSION

VITAMINS AND CAROTENOIDS

Tables 1-3 show the results of complex evaluation of patients of the first research cycle. The tables contain individual results of histological examination of prostate biopsy samples, age, as well as levels of total PSA, vitamins, and carotenoids in the blood of each patient.

Table 1 shows the results of 7 patients diagnosed with CaP based on histological examination. The Gleason score was 4 for all patients with well-differentiated CaP, 5 – with moderately differentiated CaP, and 8 – for one patient with poorly differentiated CaP. In all 7 patients with CaP (tab. 1) the concentration of vitamin A and vitamin E was not significantly different from standard values for a healthy Russian citizen [32] despite the research cycle taking place in winter and spring. Six of seven patients with CaP had lower levels of lycopene, β-carotene, and total carotenoids. Lycopene was significantly lower in four patients with well-differentiated CaP, and to a lesser extent in one patient with moderately differentiated CaP. One patient with poorly differentiated CaP (Gleason score – 8, total PSA – 100 ng/mL) did not show such a difference in micronutrient levels. Vitamin C concentration was decreased in all test subjects except for one 64-years-old patient.

The same pattern was observed in the HGPIN group (5 patients, tab. 2) – in all patients with HGPIN associated with BPH and in two patients with HGPIN associated with BPH and CP. Total carotenoid levels were decreased in 4 of 5 patients, and all five patients showed especially low lycopene concentration in the blood and percent of total carotenoids. The difference in β-carotene levels followed a less rigid pattern. No difference in vita-

min A and vitamin E was observed. It is worthy of note that one 66-years-old patient (HGPIN + BPH) with total PSA level of 5,2 ng/mL had a significantly high β-carotene concentration in the blood; total carotenoids, vitamin A, and vitamin E were close to the upper limit of the normal range. Vitamin C was insignificantly decreased in two of three patients who had undergone vitamin C level assessment.

The third group (tab. 3) – in contrast to the previous two – included nine patients who had neither cancer (tab. 1), nor premalignant lesions in the prostate (tab. 2). Therefore it can be considered as a "symbolical control group". Of nine test subjects only three patients with BPH + LGPIN had moderately lower total carotenoid and lycopene serum levels. Thus, in this "symbolical control group" changes in carotenoid concentrations are absent or mild. However, similar to the previous groups, the level of vitamin C was decreased in 5 of 7 examined patients.

In all three groups no patterns or significant changes in vitamin A and E concentrations in the blood were observed.

Tables 4 and 6 show the results of the second research cycle which included 94 males divided into the 4 aforementioned groups. The test procedures at this stage were the same as those during the first research cycle, yet in tables 4 and 6 we opted not to list all the parameters for each patient. Table 5 shows the statistical data on vitamins, LPO, and PSA, so as to assess the validity of the values showing differences among the patients from different groups.

Total PSA level in the blood serum was normal in group 4, and above 4 ng/mL in group 3 (LGPIN, BPH) and group 2 (HGPIN) ($p<0.01$). In CaP (group 1) the mean PSA concentration was maximal (16,9 ng/mL), however, there was no statistically significant difference in this parameter between CaP and HGPIN (10,4 ng/mL) groups. There are special aspects concerning the biochemical analysis results in the control groups that are worthy of note. The control group (group 4) consisting of healthy younger males with no detected prostate gland pathology and with normal PSA levels was characterized by low contents of lycopene (in 90% of cases), β-carotene (in 90% of cases), and total carotenoids (in 90% of cases) compared to normal ranges for citizens of the Russian Federation receiving adequate nutrition and recommended by the Scientific Research Institute of Nutrition [32]. This difference was statistically significant ($p<0.01$) according to the sign test [38-41]. Discussing the reasons for such findings is not among our goals, however it can be noted that the second research cycle was conducted from December to June. The other "symbolical control group" (group 3) consisted of older males with benign prostate pathology (LGPIN and/or BPH and/or CP) and with slightly elevated total PSA level within the «gray zone» (4-10 ng/mL) but no signs of prostate carcinogenesis. Their age matched the age of the patients in the other two groups (group 1 and 2). This "symbolical control group" (group 3) was also characterized by lycopene, β-carotene, and total carotenoids deficiency. However there were differences between control groups 3 and 4: younger males (group 4) had more marked β-carotene deficiency than group 3 older males. 18 of 20 group 4 males had decreased β-carotene levels ($p>0.05$ according to the Student's t-test, and $p<0.01$ according to the sign test), and 19 of 20 group 4 males had lower total carotenoid levels ($p<0.05$ according to the Student's t-test, and $p<0.01$ according to the sign test [38-41]) which might have been due to the difference in lifestyle and diet. Similar differences between the control groups in terms of microelements concentration in the blood will be discussed later. The concentration of selenium, germanium, and silicon were statistically significantly different in groups 3 and 4 (tab. 7).

Similar to the first research cycle, no statistically significant difference in vitamin A serum levels was found in all four examined groups. We only observed a slight decrease in vitamin A concentration in patients with CaP compared to other test subjects. Vitamin E (antioxidant) concentration was statistically significantly lower in patients with CaP in comparison to the control group 3 ($p<0.05$) (tab. 4 and 5).

At the same time the concentration of water soluble vitamin C (antioxidant) was lower in groups 1, 2, and 3, i.e. in patients with CaP, HGPIN, LGPIN + BPH + CP. This deficiency was statistically significant ($p<0.05$) in patients with HGPIN (fig.1, tab. 4 and 5).

LPO products are markers of intensity of peroxidation processes which may facilitate carcinogenesis. The obtained results show that increased levels of primary LPO products – conjugated dienes – were characteristic of the same three groups of patients (groups 1, 2, and 3) that had lower vitamin C levels. Elevated levels of conjugated dienes in patients with CaP, HGPIN, and benign prostate pathology (group 3) (tab. 4 and 5) were statistically significant ($p<0.01$) in comparison to control group 4. No difference in another LPO product – malonic dialdehyde – level was observed. It should be noted that this dialdehyde is a late product of oxidative degradation of unsaturated fatty acids.

Lycopene level evaluation is an essential procedure for certain studies indicate its role in CaP prevention. Lycopene is an active antioxidant that neutralizes reactive oxygen species (singlet oxygen) and free radicals. It possesses anticarcinogenic, antiatherogenic, and antimutagenic properties. Lycopene is an acyclic carotenoid (not a provitamin A), and its trans-form is contained in tomatoes and to a lesser extent in watermelons, apricots, and pink grapefruit. During heating of tomatoes the trans-isomer transforms into the cis- isomer, which absorption in the digestive tract is facilitated by the presence of fats [22, 42].

Both control groups had similar concentration of lycopene in the blood which was about 2 times lower than

the lower limit of the normal range for Russian citizens [32]. Lycopene deficiency was more marked in patients with HGPIN and even more so in patients with CaP ($p<0.05$) (fig. 2, tab. 4 and 5).

The results of the second research cycle regarding lycopene, vitamins C, A, and E corresponded well to the data obtained during the first research cycle. Other carotenoids with antioxidant properties – as mentioned earlier – were more decreased in younger males of group 4. Therefore, in this case this group could not be regarded as control for the patients of groups 1, 2, and 3. Compared to the "symbolical control group" (group 3), level of total carotenoids was somewhat smaller in patients with CaP ($p>0.05$), and statistically significantly decreased in patients with HGPIN ($p<0.05$). No difference in β-carotene level between all four groups was observed.

MICROELEMENTS

The choice of elements analyzed in our study was based on our previous experiences in the field of bioinorganic substances research [37]. However, our major consideration was searching for new data, as we had not found any literature on previous studies wherein blood concentrations of different microelements would have been measured in patients with different types of urological pathology. The general results of the analysis of 21 chemical elements in the blood are shown in table 6. Statistically significant differences according to Student's t-test [38-40] were observed in aluminum, germanium, sulfur, selenium, and some other elements (tab.6 and 7).

Aluminum. Aluminum concentration in the first group (CaP) was over 10 times more than in control group 4. The difference was statistically significant ($p<0.01$). Aluminum concentration was higher in group 2 (HGPIN) than in group 1. However, this difference was not statistically significant ($p>0.05$). Aluminum concentration in group 2 (HGPIN) was statistically significantly higher than in control group 3 ($p<0.05$) and control group 4 ($p<0.01$). Aluminum level in patients with precancer (HGPIN) was three times higher than in patients with BPH (group 3) and 15 times higher than in control group 4. The difference in aluminum concentration between the third and fourth control groups was not statistically significant, although the level was higher in the third group (fig. 4, tab. 6 and 7).

Thus, aluminum concentration in the blood was 15 times higher in HGPIN and 10 times higher in CaP.

A high concentration of aluminum is known to possess pro-oxidant properties: it activates LPO of phospholipids and damages membranes of hepatocytes [43], modeled membranes [44] and membranes of the nervous tissue facilitating neurodegeneration in Parkinson's disease and Alzheimer's disease thus impairing memory [45,46]. Aluminum is also capable of accumulation in rat erythrocytes which intensifies LPO and causes hemolysis [47]. Aluminum is believed to have several mechanisms of toxicity: it increases the formation of reactive oxygen species induced by reduced Fe^{2+} ions [43-48] and inhibits the activity of a number of enzymes involved in antioxidant defense by directly influencing the enzyme molecules or their synthesis (it competes with magnesium as an enzymatic cofactor [49]) and by suppressing the antioxidant effects of flavonoids [43]. Normal and excessive aluminum concentration in the body results from intake from water, food (vegetables, rice, wheat, etc.), dust-filled air, drugs and damaged aluminum dishware, as well as from deodorants and cosmetic substances. The routes of entry through which aluminum gains access into the human body are gastrointestinal tract, lungs, and skin. Excessive toxic aluminum is accumulated in the liver, bones and brain. At the same time aluminum as a metal is essential for the development of the intercellular matrix, connective tissue of the skeletal bone and cartilage as well as for iron metabolism regulation. The main (and, possibly, the only) source of aluminum for all living organisms is soil [37].

The obtained results are one of the first to show that aluminum is involved in prostate carcinogenesis.

Germanium. The highest concentration of germanium was observed in control group 4, the lowest – in cancer and precancer groups (groups 1 and 2). Germanium level was statistically significantly lower in group 1 than in group 4 ($p<0.01$), as well as in group 2 ($p<0.01$) and group 3 ($p<0.05$) in comparison to group 4. Germanium concentration was slightly lower in patients with precancer (HGPIN) than with CaP, however, the difference was not statistically significant ($p>0.05$). The difference between group 2 and group 3 was also not statistically significant.

Taking into consideration the aforementioned results we can observe the following pattern: germanium concentration was higher in control groups 3 and 4, and statistically significantly lower in prostate cancer and precancer ($p<0.01$).

Germanium is well-known to possess multiple biological properties: antioxidant, anticarcinogenic, radioprotector, and immunostimulant. Entering the human body primarily with food (garlic, fish, tomatoes, beans, cepes etc.), it is exposed to bacterial enzymes in the digestive tract and transforms into organic metalloid, similar to some other microelements [50]. Several drugs have been developed [51-53] – one of them being bis-carboxyethyl germanium sesquioxide (Ge 132) – which use is recommended in patients with various diseases, including pathologies involving free radical damage. Germanium, germanium dioxide, and its various organic forms with anticarcinogenic and antioxidant properties stimulate main enzymes involved in the antioxidant defense in the tissues, decrease the quantity of free radicals, and inhibit LPO [51-54]. For example, Ge 132 blocks LPO in mouse liver by activating antioxidant enzymes – superoxide dismutase and catalase [55]. Precancerous lesions of rat oral cavity induced by methylnitrosoguanidine undergo regression during combined therapy with Ge 132 and selenium yeast [56].

Table 1.

Examination results of patients with CaP (1st cycle)

Age, years	Parameters	Concentration in the blood								
		Total PSA – ng/mL	Vit.C, mg/dL	Vit.A, µg/dL	Vit.E, mg/dL	Lycopene, µg/dL	β-carotene, µg/dL	Total carotenoids µg/dL	% of total carotenoids	
									lycopene	β-carotene
64	CaP, well-differentiated	9.8	1.10	56.0	0.75	6.2	12.3	60.1	10	20
67	CaP, well-differentiated	19.0	-	63.2	1.48	14.1	14.8	76.7	18	19
77	CaP, well-differentiated	100,0	0.49	48.4	1.68	1.8	10.0	23.9	8	42
75	CaP, well-differentiated	8.1	0.44	72.4	1.11	8.2	16.3	49.6	16	32
73	CaP, well-differentiated	15.5	0.60	57.9	0.73	0.5	5.3	17.9	3	30
66	CaP, moderately differentiated	7.1	0.39	61.6	0.98	12.1	12.0	42.8	28	28
68	CaP, poorly differentiated	100,0	0.25	52.2	0.93	27.8	22.1	85.3	33	26
	Normal values	⩽4*	⩾0.7	30-80	0.8-1.5	22.0-35.0	20-40	80-230	19	20

Note. In foreign countries normal PSA levels are age-specific: 40 to 49 years – 2.5; 50 to 59 years – 3.5; 60 to 69 years – 4.5; 70 years and older – 6.5 ng/mL [52]

Table 2.

Examination results of patients diagnosed with HGPIN based on histological examination (1st cycle)

Age, years	Parameters	Concentration in the blood								
		Total PSA – ng/mL	Vit.C, mg/dL	Vit.A, µg/dL	Vit.E, mg/dL	Lycopene, µg/dL	β-carotene, µg/dL	Total carotenoids µg/dL	% of total carotenoids	
									lycopene	β-carotene
77	HGPIN + BPH + CP	13.0	0.63	50.5	1.00	1.1	16.2	53.7	2	30
63	HGPIN + BPH + CP	19.8	0.60	82.4	1.92	3.0	48.7	77.1	4	63
63	HGPIN + BPH	6.8	1.02	82.2	1.40	2.9	16.2	31.5	9	51
63	HGPIN + BPH	4.0	-	60.3	0.99	2.9	7.6	30.9	9	25
66	HGPIN + BPH	5.2	-	80.6	1.36	8.1	112.1	212.5	4	52
	Normal values	⩽4	⩾0.7	30-80	0.8-1.5	22.0-35.0	20-40	80-230	19	20

Table 3.

Examination results of patients with BPH based on histological examination including cases of BPH associated with LGPIN and/or CP (1st cycle)

Age, years	Parameters	Concentration in the blood								
		Total PSA – ng/mL	Vit.C, mg/dL	Vit.A, µg/dL	Vit.E, mg/dL	Lycopene, µg/dL	β-carotene, µg/dL	Total carotenoids µg/dL	% of total carotenoids	
									lycopene	β-carotene
60	BPH+LGPIN+CP	14.7	0.63	64.0	1.37	6.1	17.2	51.5	12	33
72	BPH+LGPIN+CP	6.0	0.46	63.2	1.17	8.9	22.6	54.6	16	42
51		2.2	1.34	57.3	1.08	6.8	15.1	68.3	10	22
65	BPH+CP	BPH+LGPIN 26.4	-	73.0	1.27	13.8	24.0	72.1	19	33
62	BPH+CP	11.7	0.21	67.4	1.21	28.3	12.9	55.6	50	23
66	BPH+CP	7.7	0.32	51.3	0.67	16.5	165.9	220.8	7	75
77	BPH+CP	11.2	0.77	63.5	1.08	36.6	76.7	181.5	20	43
75	BPH	15.2	0.14	49.6	0.99	7.5	16.4	61.4	12	27
54	BPH	5.6	-	78.7	0.99	13.0	9.9	41.6	31	24
	Normal values	⩽4	⩾0.7	30-80	0.8-1.5	22.0-35.0	20-40	80-230	19	20

Table 4.

Concentration of vitamins, carotenoids, and lipid peroxidation products in patients with various prostate pathologies and morphological changes of the prostate gland

N – number of patients in a group, X – mean value, σx^{-} – standard error of mean (mean error)

Parameters. Normal values are given in parentheses	Patient groups							
	Group 1 CaP		Group 2 HGPIN		Group 3 LGPIN, BPH, CP		Group 4 Control	
	$X \pm \sigma x^{-}$	N	$X \pm \sigma x^{-}$	N	$X \pm \sigma x^{-}$	N	$X \pm \sigma x^{-}$	N
Vitamin A µg/dL (30-80)	60.4±3.1	26	66,9±3.8	26	65.4±3.1	22	68.2±3.8	20
Vitamin E mg/dL (0.8-1.5)	1.13±0.06	25	1.25±0.10	26	1.36±0.09	23	1.20±0.10	20
Lycopene µg/dL (22-35)	+) 5.16±0.82	25	+) 6.87±1.12	25	10.57±2.10	23	11.91±2.68	20
β-carotene µg/dL (20-40)	17.2±3.0	26	12.6±1.4	26	17.2±3.2	23	10.9±1.5	20
Total carotenoids µg/dL (80-230)	53.0±6.0	26	+) 44.8±4.5	25	+) 65.3±8.4	21	45.0±5.5	20
Vitamin C mg/dL (⩾0.7)	0.560±0.069	26	0.475±0.052	26	0.542±0.081	23	0.700±0.090	20
conjugated dienes units of optical density	0.159±0.006	23	0.155±0.005	25	0.161±0.005	21	0.122±0.009	19
Malonic dialdehyde nmole/mL	7.03±0.22	23	7.01±0.22	26	7.90±0.40	21	7.61±0.32	19
Total PSA ng/mL	16,9 ± 4,0	26	10,4 ± 1,4	26	6,3 ± 1,0	23	1,4 ± 0,4	20

Note. In cases marked with a « +)» symbol in accordance with "The rule of excluding the so-called outlying cases" (by V.I. Romanovsky for a probability of 99%) [32], one variant was excluded

Table 5.

t-values (Student's t-test) obtained as a result of pairwise comparison of concentrations of each component between various groups of patients (groups 1-4)

	Compared patient groups					
Parameters	1-2	1-3	1-4	2-3	2-4	3-4
Vitamin A	1.33	1.14	1.59	0.31	0.24	0.57
Vitamin E	1.03	2.13	0.60	0.82	0.35	1.19
Lycopene	1.23	2.40	2.41	1.55	1.73	0.39
β-carotene	1.39	0	1.88	1.32	0.83	1.78
Total carotenoids	1.09	1.19	0.98	2.15	0.03	2.03
Vitamin C	0.98	0.17	1.23	0.69	2.17	1.31
Conjugated dienes	0.51	0.26	3.42	0.84	3.23	3.82
Malonic dialdehyde	0.06	1.89	1.49	1.93	1.53	0.57
Total PSA	1,53	2,57	3,85	2,38	6,16	4,54

Note. Statistically significant differences (in bold) with the number of degrees of freedom being $N_a+N_b-2=50$ (40): $t > 2.01$ (2.02) corresponds to $p<0.05$; $t >2.68$ (2.70) corresponds to $p<0.01$ [31]

Table 6.

Concentration of chemical elements in the blood (μg/mL) in various prostate pathologies and morphological changes of the prostate gland. N – number of patients in a group, X – mean value, $\sigma\bar{x}$ – standard error of mean (mean error)

Chemical element	Group №1 CaP		Group №2 HGPIN		Group №3 LGPIN,BPH,CP		Group №4 CONTROL	
	$X \pm \sigma\bar{x}$	N	$X \pm \sigma\bar{x}$	N	$X \pm \sigma\bar{x}$	N	$X \pm \sigma\bar{x}$	N
Se x 10^{-}I	1.20±0.09	26	1.20±0.05	25	1.70±0.05	21	1.11±0.08	20
Ca	48.1±1.84	26	49.7±2.65	25	52.8±3.29	21	52.9±3.67	20
Pb x 10^{-2}	4.61±0.80	26	5.17±0.82	25	4.30±1.03	21	3.52±0.89	20
S	332±9	26	316±11	25	352±30	21	357±10	20
Cu x 10^{-1}	4.54±0.36	26	4.04±0.33	25	4.55±0.37	21	4.61±0.33	20
Fe	264±9	26	251±6	25	265±8	21	273±9	20
Si x 10^{-1}	7.32±3.20	26	5.75±1.73	25	2.94±1.29	21	9.30±2.02	20
Sr x 10^{-3}	6.97±2.39	26	7.08±5.80	25	6.55±1.60	21	4.70±1.38	20
Co x 10^{-3}	1.99±0.95	26	1.02±0.16	25	1.16±0.14	21	1.14±0.20	20
Al x 10^{-2}	1.16±0.32	26	1.65±0.39	25	0.56±0.29	21	0.11±0.02	20
Mo x 10^{-}4	12.17±5.5	26	7.30±2.70	25	4.13±0.40	21	4.73±0.37	20
Mn x 10^{-}4	3.75±0.76	26	12.25±6.76	25	1.92±0.62	21	2.88±1.37	20
Ba x 10^{-}4	0.81±0.25	26	2.50±1.70	25	1.48±0.75	21	0.16±0.05	20
Cr x 10^{-}6	5±2	26	5±1	25	3±1	21	3±1	20
Ge x 10^{-3}	2.80±0.17	26	2.60±0.16	25	2.91±0.14	21	3.53±0.20	20
K	833±43	26	732±26	25	798±20	21	849±31	20
Na	1421±61	26	1345±74	25	1402±58	21	1395±71	20
Li x 10^{-}4	5.61±1.45	26	4.94±0.76	25	7.38±2.93	21	3.88±1.27	20
Ti x 10^{-2}	2.77±1.41	26	3.31±1.78	21	9.34±6.68	21	0.34±0.11	20
P	113±6	26	108±6	25	111±4	21	122±4	20
Zn	3.68±0.19	26	3.77±0.20	25	3.55±0.16	21	4.06±0.36	20

Table 7.

t-values (Student's t-test) obtained as a result of pairwise comparison of concentrations of each component between various groups of patients (groups 1-4)

Chemical element	Compared patient groups					
	1-2	1-3	1-4	2-3	2-4	3-4
Se	0	**4.85**	0.74	**7.07**	0.95	**6.25**
Ca	0.49	1.24	1.16	0.73	0.70	0.02
Pb	0.48	0.23	0.91	0.66	1.36	0.57
S	1.11	0.63	1.83	1.12	**2.76**	0.15
Cu	1.02	0.02	0.14	1.02	1.22	0.12
Fe	1.20	0.08	0.70	1.40	**2.03**	0.66
Si	0.43	1.26	0.52	1.30	1.33	**2.65**
Sr	0.02	0.14	0.82	0.08	0.39	0.87
Co	1.00	0.86	0.87	0.65	0.46	0.08
Al	0.97	1.38	**3.27**	**2.24**	**3.94**	1.54
Mo	0.79	1.45	1.34	1.16	0.94	1.10
Mn	1.24	1.86	0.55	1.52	1.35	0.63
Ba	0.98	0.84	**2.54**	0.54	1.37	1.75
Cr	0	0.89	0.89	1.41	1.41	0
Ge	0.85	0.49	**2.78**	1.45	**3.63**	**2.54**
K	2.00	0.73	0.30	2.01	**2.89**	1.38
Na	0.79	0.22	0.27	0.60	0.48	0.07
Li	0.40	0.54	0.89	0.80	0.71	1.09
Ti	0.23	0.96	1.71	0.87	1.66	1.34
P	0.58	0.27	1.24	0.41	1.94	1.94
Zn	0.32	0.52	0.93	0.85	0.70	1.29

Note. Statistically significant differences (in bold) with the number of degrees of freedom being $N_a+N_b-2=50$: $t > 2.01$ corresponds to $p<0.05$; $t > 2.68$ corresponds to $p<0.01$ [31]

The results of our studies regarding the decrease of germanium levels in the human body during prostate carcinogenesis correspond to the aforementioned published findings.

Sulfur. We observed lower concentrations of sulfur in prostate cancer and precancer groups than in control groups (fig. 5, tab.6 and 7). However, a statistically significant difference was only found between the precancer group (HGPIN) and control group 4 ($p<0.01$). Sulfur is an important part of the human antioxidant system: mainly cysteine and its derivative –glutathione, CoA-SH coenzyme, and corresponding enzymes. For example, glutathione peroxidase facilitates hydrogen peroxide and fatty acid hydroperoxide degradation thus blocking LPO. Reduced glutathione is a coenzyme for this enzyme.

Our results suggest that inadequate intake of sulfur-containing substances and lower concentrations of sulfur (as well as cysteine, coenzymes and enzymes) in tissues may be one of the factors of prostate epithelium carcinogenesis.

Selenium. We observed nearly identical selenium concentration in the blood of the patients of groups 1, 2, and 4, meaning that patients with precancer and cancer had nearly the same level of selenium in the blood as healthy younger males (fig. 6, tab. 6 and 7). If we take into consideration the widely accepted theory about selenium (healthy dietary intake level) being an effective antioxidant, interpretation of the obtained data proves difficult at first sight. It should be noted, however, that all examined patients had a diet low in selenium: our results showed a mean concentration (tab. 6) of 11-17 μg/L in the serum. The lower limit of the normal range is 60-80 μg/L [57-58]. This fact may have an influence on the interpretation of the obtained data. That is why it should be pointed out that group 1 and group 2 (CaP and HGPIN) patients had a statistically significantly lower level of selenium ($p<0.01$) than patients of the "symbolical control group" – group 3 (BHP, LGPIN, CP) – who showed no signs of prostate carcinogenesis. The age of group 3 patients corresponded to the age of group 1 and group 2 patients who were older than males of the fourth control group. If we include this into the analysis, selenium deficiency (as an antioxidant) in CaP and HGPIN (groups 1 and 2) seems plausible. The inadequacy of group 4 as a control group in this case is supported by the fact that selenium concentration in the blood in the third "symbolical control group" (BPH, etc.) was statistically significantly higher ($p<0.01$) than in the fourth control group of younger males (fig.6, tab. 6 and 7).

Similarly, we observed several differences in the concentration of other microelements between two control groups: silicon (Si) and germanium (Ge) levels were statistically significantly lower in group 3 ($p<0.05$) than in group 4 (tab. 6 and 7).

The results also show certain differences regarding other 4 chemical elements between various patient groups. All patients with uropathology (groups 1, 2, and 3) had lower concentrations of iron, silicon, and potassium in the blood and a higher concentration of barium in comparison to healthy males (group 4). Statistically significant changes in the concentration of the following elements were found: iron ($p<0.05$) and potassium ($p<0.01$) in group 2 (HGPIN), silicon ($p<0.05$) in group 3 (BPH, etc.), and barium ($p<0.05$) in group 1 (CaP) (tab. 6 and 7). The aforementioned results are merely given as facts without any attempts at discussion and interpretation.

Therefore, according to our data, changes in the level of four chemical elements (aluminum, germanium, sulfur, and selenium) in the blood were already observed in patients with precancerous changes of the prostate. In order to give greater credibility to such statements further research is certainly required, namely microelement concentration assessment in prostatic tissue or even prostate epitheliocytes. The difference in mineral concentration in the blood may be due to aspects of diet, lifestyle, gastrointestinal tract status, since the majority of microelements enter the organism by mouth (per os). For this reason we discussed the list of microelement sources (aluminum and germanium) for humans above.

The results concerning other 17 elements in four patient groups given in table 6 might prove useful as reference material for further research. It should be noted that according to the current non-official classification, to with we do not generally adhere, these chemicals can be divided into micro- and macroelements. We have to acknowledge that, in accordance with this classification, the blood concentration of some of the macroelements (Ca, S, Fe, K, Na, P) studied is indeed somewhat higher (table 6), although it does not exceed 1 mg/mL of human blood (except for sodium).

DISCUSSION

It is essential to know how certain tumor markers change during cancer initiation, promotion, formation, progression, and even metastasis. In our previous works we attempted to follow this strategy of CaP research for we had an opportunity to monitor the same markers during the different stages of prostate diseases: BPH, CP, LGPIN, HGPIN (precancer), localized CaP, metastatic CaP. We analyzed multiple clinical parameters, results of histological examination, the Gleason sum in cancer biopsy specimens or tissues obtained during surgery, DNA ploidy, telomerase activity, PSA, microelements, vitamins, carotenoids, LPO products. We also detected early micrometastases by assessing the blood for prostatic cells containing mRNA which codes for PSA [26-31,59,60]. With the results given in the present article, we can see the following trends in the changes of several biochemical parameters in the blood of patients with urologic oncopathology.

A. CaP is characterized by decreased levels of lycopene, vitamin E, vitamin A, germanium (all of them be-

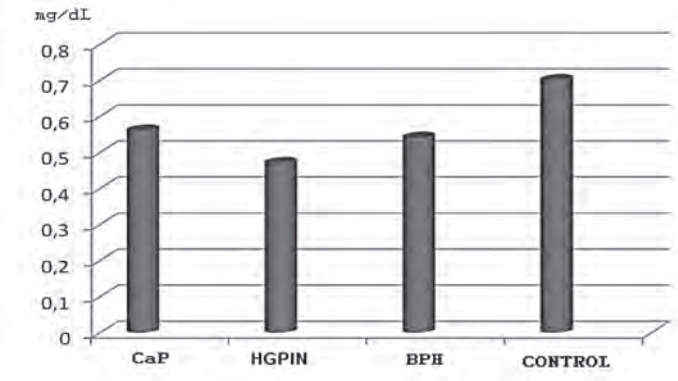

Fig. 1. Blood serum vitamin C levels (mg/dL)

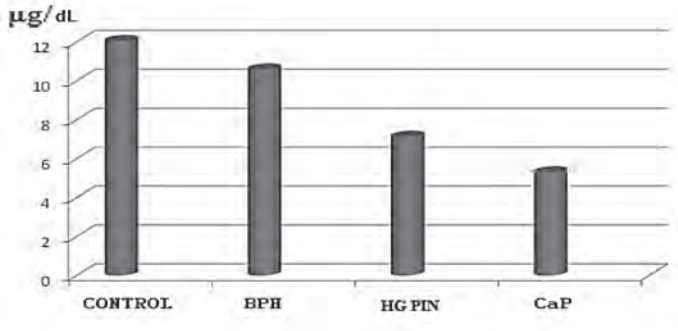

Fig. 2. Blood serum lycopene levels (µg/dL)

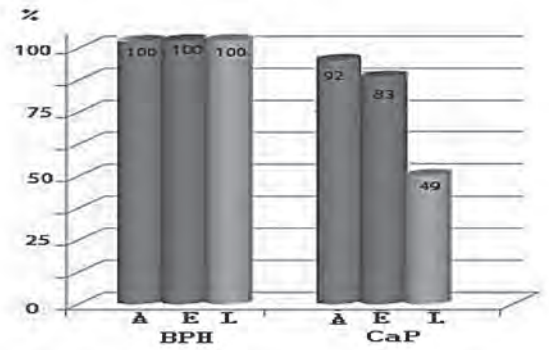

Fig. 3. Differences in vitamin A (A), vitamin E (E) and lycopene (L) concentration (%) in the blood serum between group 1 (CaP) and group 3 (BPH, LGPIN, CP)

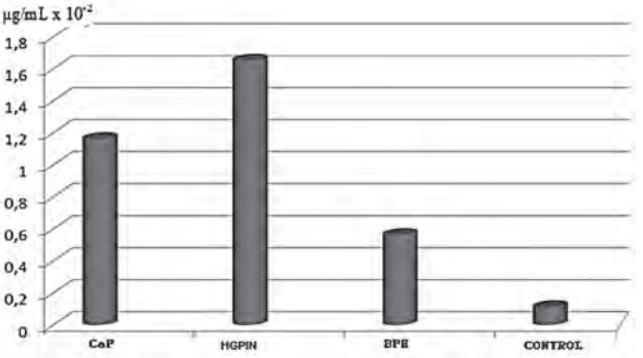

Fig. 4. Blood aluminum levels (µg/mL×10^{-2})

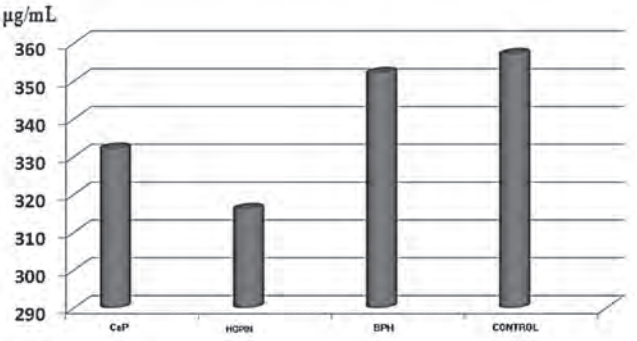

Fig. 5. Blood sulfur levels (µg/mL)

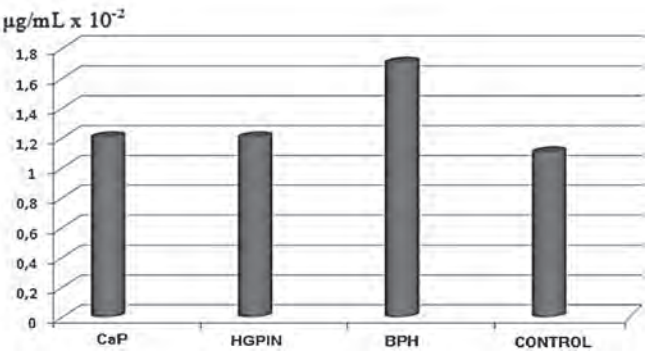

Fig. 6. Blood selenium levels (µg/mL×10^{-2})

ing antioxidants) and increased levels of pro-oxidants – aluminum and LPO products (conjugated dienes) in comparison to other carcinogenesis stages or prostate pathologies (LGPIN, BPH, CP) or healthy individuals. Figure 3, tables 4 and 6 demonstrate the mentioned changes in CaP.

B. High-grade prostatic intraepithelial neoplasia (HGPIN), which can be considered as precancer according to multiple sources [12-16], is characterized by decreased levels of antioxidants (vitamin C, total carotenoids, lycopene, germanium, sulfur, selenium) and increased levels of pro-oxidants (aluminum and conjugated dienes) in comparison to other carcinogenesis stages or prostate pathologies (LGPIN, BPH, CP) or healthy individuals.

The aforementioned statements may be regarded as trends that require further proof for although the differences were proven to be statistically significant in the majority of cases, changes in certain parameters were not found to be statistically significant.

The data presented in the article may have potential clinical implications. Firstly – as mentioned earlier in the introduction – some urologists have recently changed their views on HGPIN. Having diagnosed HGPIN based on histological examination, physicians and scientists consider shifting from passive monitoring of these patients to active therapeutic strategies in order to postpone and/or stop the transformation of HGPIN into CaP [12-20]. Therefore, our results concerning the role of anti- and pro-oxidants in prostate carcinogenesis may

help further research of this subject and generally provide support for and increase the importance of other works devoted to CaP epidemiology and prophylaxis. Secondly, the results presented are of certain scientific interest for they contribute experimental data to the understanding of the pathogenic role of anti- and pro-oxidants in CaP initiation, development, and formation.

Our study was approved by an independent local ethics committee. The patients gave their informed consent to the participation in the study, within the framework of which some of their blood samples (taken as part of routine laboratory testing) were also used to collect data for the study.

CONCLUSIONS

1. The results showed statistically significant decreased blood levels of antioxidants (lycopene, vitamin E, germanium, selenium) and increased levels of pro-oxidants – lipid peroxidation products (conjugated dienes) and aluminum – in patients with prostate cancer (CaP).

2. No statistically significant difference in blood levels of the studied vitamins, vitamin-like substances, lipid peroxidation products, and microelements was found between the patients with high-grade prostatic intraepithelial neoplasia (HGPIN) and CaP. In patients with HGPIN (precancer) aluminum level increase was more marked (15 times higher) than in patients with CaP (10 times higher) in comparison to patients with a healthy prostate gland.

3. Patients with HGPIN had decreased blood levels of vitamin C, total carotenoids, germanium, selenium and increased levels of conjugated dienes and aluminum in comparison to the control groups.

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