# Innovative Methods of Elementosis Study in Oncourological Practice

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**ABSTRACT**

The purpose of this work is to study the content of macronutrients in the tissues with oncourological pathology. Methods: Clinical examination material of 279 people was used in the work, 229 of them were men (82%) and 50 were women (18%). Elemental analysis of oxygen, carbon, calcium, nitrogen and sulfur was carried out using a detector to record the spectra of characteristic X-ray radiation (EPAX company), which were integrated with "Quanta 600 FEG" scanning electron microscope. Results: with prostate cancer, the oxygen content decreased, so during stage 1, the oxygen content decreased by 36.8% among middle-aged patients, and by 38.6% among elderly patients, stage 2 - by 32.4% and 28.9%, stage 3 - by 34.1% and 34.2%, and stage 4 - by 30.9% and 35.1%. The nitrogen content changed insignificantly, carbon and sulfur decreased. The calcium index among middle-aged patients with stage 1 prostate cancer increases by 10.6, and by 10.8 times among the elderly, while it is absent among the patients with stage 2, 3 and 4. The nitrogen content among the patients with renal pathology did not change significantly, but there was a tendency of carbon, calcium, and sulfur increase and oxygen decrease. When they studied the level of macronutrients in bladder cancer, there was a tendency to nitrogen and carbon level increase, and in the groups of stage 1 and 2 patients, the content of calcium and sulfur increased by 12.5 and 3.8 times, respectively, and oxygen was also reduced. Conclusions: we found that all groups demonstrated oxygen content decrease, most pronounced among stage 2 patients with bladder cancer - 49.5%, which leads to tissue hypoxia in the studied organs. The nitrogen and carbon content varied slightly. The content of calcium and sulfur increases among the patients of all studied groups.

**Keywords:** oncourology, trace elements, prostate, kidneys, bladder



## INTRODUCTION

The body of a healthy person has a clear self-regulating system of homeostasis, in which chemical elements play an important role. Their level in the blood and body tissues is subject to certain physiological patterns. Elemental homeostasis is a particular form of the general homeostatic body system, the violation of which affects the body ability to adapt in extreme conditions (Chernova 2018; Kanzhigalina et al. 2013; Pavlova et al. 2019).

The stability of the chemical composition is one of the most important and indispensable conditions for the normal functioning of the body. The kinetics, distribution and deposition of metal ions is subject to the biochemical regulation of the macroorganism. The change of each of the macro-micronutrients concentration is interconnected. Therefore, both the deficiency of macro- and microelements, as well as their increased concentration, can lead to adverse consequences for human life (Chernova 2018; Kanzhigalina et al. 2013; Pavlova et al. 2019).

In Russia, kidney cancer is the first one in the structure of the urinary tract oncological pathology and accounts for 2.7% of all malignant neoplasms among adults. According to the rate of average annual growth in Russia, this tumor occupies the third place (Zolotukhin et al. 2016; Pavlova et al. 2019; Raaschou-Nielsen et al. 2017; Pavlova et al. 2016).

Due to the constant increase of morbidity and mortality, prostate cancer is one of the urgent problems of oncourology in Russia (Alekseev et al. 2016; Kaprin et al. 2018; Buevich et al. 2019). This pathology is more common among middle-aged and elderly men, and takes the 2nd place in the structure of cancer incidence among men, accounting for 14.5%. With age, the risk of this disease development increases by 3-4% per year (Kaprin et al. 2018). So, in 2008, 60 cases of the disease were registered in Russia per 100 thousand people, and 162.2 in 2018 (Kaprin et al. 2018; Pourmousa et al. 2018; Tel H, Ertekin Pinar & Daglar 2018).

In the structure of oncological morbidity, bladder tumors make up from 2 to 5% of all neoplasms. Every year, 335.8 thousand people become ill with bladder cancer and 132.4 thousand die in the world, that is, one of three dies from this serious disease. Bladder tumors among men occur 3-4 times more often than among women. The increase of patients with bladder cancer in Russia made 8.3%, rising in relative numbers from 8.9 to 9.7 per 100,000 of population. It should be noted that at present only 45% of patients have an early diagnosis of bladder cancer (Khudyashev & Kaprin 2010; Dugué et al. 2018).

Thus, the social significance of this pathology is so great that timely diagnosis of tumors and cancer patient treatment remain an urgent problem of modern oncology.

In this regard, the aim of our study was to study the content of macroelements in the tissues with oncourological pathology.

## MATERIALS AND METHODS

The work was based on material received from 2013 to 2018 at the bases of the Belgorod Oncology Center, Belgorod Regional Clinical Hospital of St. Joseph, and the Belgorod Pathological Bureau. The study of the material, analysis and processing of the obtained results was carried out at the Department of Pathology and at the Belgorod State University, scientific and educational and innovation center "Nanostructured materials and nanotechnologies".

279 people were studied within the work, of which 229 were men (82%) and 50 were women (18%). Groups were formed according to the age and nosological criteria (tab. 1, 2).

**Table 1: Men suffering from prostate diseases**

| Control | | Men suffering from prostate diseases (n=115) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Prostate cancer | | | | | | | |
| Middle age (40-49 years) | Elderly age (60-83 years) | Benign hyperplasia prostate (Elderly age, 60-77 years) | Middle age n=58 (40-49 years) | | | | Elderly age n=42 (61-79 years) | | | |
| | | | I stage ($T_1$ $N_0$ $M_0$) | II stage ($T_1$-$T_2$ $N_0M_0$) | III stage ($T_1$-$T_3$ $N_1$-$N_2$) | IV stage ($T_1$-$T_3$ $N_1$-$N_2$ $M_1$) | I stage ($T_1$ $N_0$ $M_0$) | II stage ($T_1$-$T_2$ $N_0M_0$) | III stage ($T_1$-$T_3$ $N_1$-$N_2$) | IVstage ($T_1$-$T_3$ $N_1$-$N_2$ $M_1$) |
| n=10 | n=10 | n=15 | n=15 | n=14 | n=14 | n=15 | n=10 | n=15 | n=11 | n=6 |

**Table 2: Patients with pathology of the kidneys and bladder**

| | | | | |
|---|---|---|---|---|
| Pathology of the kidneys, bladder (n=144) | Control (n=20) | Middle age (40-49 years) | | n=10 |
| | | Elderly age (60-83 years) | | n=10 |
| | Kidney cysts (n=22) | Middle age (41-55 years) | | n=10 |
| | | Elderly age (60-78 years) | | n=12 |
| | Kidney cancer (women 40 and men 52) (n=92) | Middle age (40-55 years) (n=43) | I stage ($T_1$ $N_0$ $M_0$) | n=10 |
| | | | II stage ($T_1$-$T_2$ $N_0M_0$) | n=12 |
| | | | III stage ($T_1$-$T_3$ $N_1$-$N_2$) | n=13 |
| | | | IV stage ($T_1$-$T_3$ $N_1$- $N_2$ $M_1$) | n=8 |
| | | Elderly age (61-79 years) (n=49) | I stage ($T_1$ $N_0$ $M_0$) | n=10 |
| | | | II stage ($T_1$-$T_2$ $N_0M_0$) | n=18 |
| | | | III stage ($T_1$-$T_3$ $N_1$-$N_2$) | n=16 |
| | | | IV stage ($T_1$-$T_3$ $N_1$- $N_2$ $M_1$ | n=5 |
| | Bladder cancer (women 10 and men 20) | Elderly age (60-71 years) (n=30) | I stage ($T_1$ $N_0$ $M_0$) | n=5 |
| | | | II stage ($T_1$-$T_2$ $N_0M_0$) | n=15 |
| | | | IIIstage ($T_1$-$T_3$ $N_1$-$N_2$) | n=6 |
| | | | IV stage ($T_1$-$T_3$ $N_1$- $N_2$ $M_1$) | n=4 |

All subjects did not have chronic forms of diseases in the acute stage, as well as severe concomitant somatic pathology. Also, the patients of the control groups did not show complaints of urological nature, and did not specifically address the experts of this profile.

For histological examination under light microscopy, samples were excised from various parts of the prostate gland, kidneys and bladder, which were fixed, embedded in paraffin and sections were prepared on a microtome, followed by their staining with hematoxylin and eosin, then they were studied and photographed using the light microscope "Topic-T" Ceti.

For scanning electron microscopy, samples were taken without fixation; this made it possible to study them without structural changes in the process of fixation. Macroelement analysis was performed using a detector to record the characteristic x-ray spectra of EPAX company. The detector is integrated with "FE1 Quanta 600 FEG" scanning electron microscope. We have studied the following macronutrients: calcium, nitrogen, carbon, oxygen and sulfur.

Statistical analysis of the data was carried out using standard methods of mathematical and statistical processing and the software MS OfficeExcel and Statistica 6.0.

In order to identify somatic pathology, diagnostic measures were taken: collection of complaints and anamnesis with a focused survey on systems and organs, physical examination, as well as laboratory and instrumental methods of the study: general blood test, biochemical general therapeutic blood

test, general urinalysis, electrocardiogram registration, study respiratory function, and chest x-ray.

If patients have pathologies of the prostate gland, kidneys and bladder, a laboratory and instrumental examination was performed in order to make and clarify the diagnosis: a comprehensive ultrasound examination of the internal organs, lymph nodes, microbiological examination of urine, skeleton bone scintigraphy, and computed tomography, if necessary. The study included the patients with histological verification of the disease.

## RESULTS AND DISCUSSION

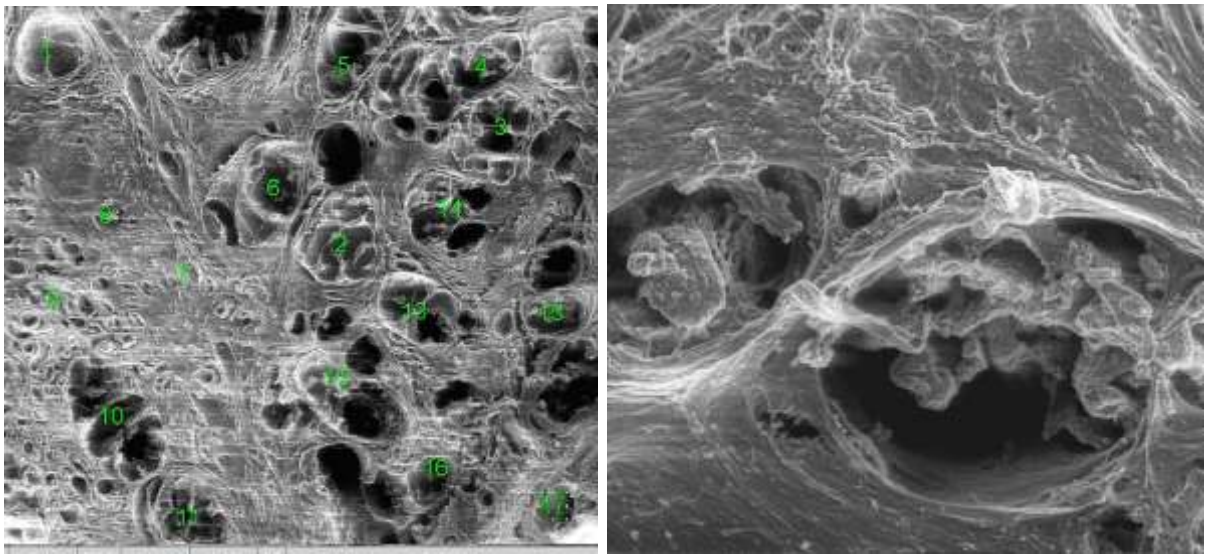
As the result of the study, it was found that the oxygen content among the control group men was the following: 21.25 ± 1.78% among middle-aged, and 20.21 ± 1.87% among elderly from the total composition of the studied components, decreasing by 23% with benign prostatic hyperplasia (15.53 ± 1.49 %). We found that in comparison with the control group, the oxygen content with prostate cancer was significantly reduced. So at stage 1, it reduced by 36.8% among middle-aged patients, and by 38.6% among elderly, stage 2 - by 32.4% and 28.9%, stage 3 - by 34.1% and 34.2%, and stage 4 - by 30.9% and 35.1% (Table 3).

**Table 3: The ratio of macronutrients in patients with prostate pathology**

| Ratio of macronutrients (%) | | | O | N | C | Ca | S |
|---|---|---|---|---|---|---|---|
| Control | age | Middle (n=10) | 21.25±1.78 | 9.71±1.22 | 67.97±2.13 | 0.14±0.01 | 0.42±0.04 |
| | | Elderly (n=10) | 20.21±1.87 | 9.91±1.31 | 68.98±2.14 | 0.12±0.02 | 0.45±0.04 |
| Benign hyperplasia prostate | age | Elderly (n=15) | 15.53±1.49 | 10.13±1.27 | 72.13±1.31 | 0.10±0.03 | 0.65±004 |
| Prostate cancer stage 1 ($T_1$ $N_0M_0$) | age | Middle (n=15) | 13.42±1.52* | 9.08±1.31 | 73.69±2.41 | 1.49±0.03 | 0.71±0.03* |
| | | Elderly (n=10) | 12.40±1.43* | 9.69±1.22 | 74.88±2.31 | 1.30±0.05 | 0.72±0.04* |
| Prostate cancer stage 2 ($T_1$-$T_2$ $N_0M_0$) | age | Middle (n=14) | 14.36±1,35* | 9.08±1.31 | 71.45±3.11 | 0 | 0.85±0.04* |
| | | Elderly (n=15) | 14.36±1.36* | 9.69±1.22 | 71.56±2.41 | 0 | 0.85±0.03* |
| Prostate cancer stage 3 ($T_1$-$T_2$-$T_3$ $N_1$- $N_2$ $M_0$) | age | Middle (n=14) | 13.99±152* | 9.41±143 | 69.57±1.98 | 0 | 0.95±0.01* |
| | | Elderly (n=11) | 13.29±2.10* | 9.50±1.65 | 70.48±1.65 | 0 | 0.90±0.01* |
| Prostate cancer stage 4 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_1$) | age | Middle (n=15) | 14.67±1.56* | 9.60±1.32 | 71.24±2.31 | 0 | 0.83±0.03* |
| | | Elderly (n=6) | 13.10±1.28* | 10.09±1.06 | 73.49±2.51 | 0 | 0.93±0.02* |

*$p<0.05$ with respect to the control group

The nitrogen content among the patients with prostate pathology did not change significantly as compared with the control group, but there was the tendency to carbon and sulfur decrease. The calcium index in the group of middle-aged patients with stage 1 prostate cancer increases by 10.6, and by 10.8 times among the elderly, and it is absent among the patients with the stage 2, 3, and 4.



A B

**Figure 1: A fragment of the prostate in organ cancer.Stage 2 ($T_1$-$T_2$ $N_0$ $M_0$). Man, 61 years old. Papillary cancer. 1-17 - places for the determination of elements (A). Follicular lesions Fig. B (x1000) fragment of Fig. A (x100). SEM.**

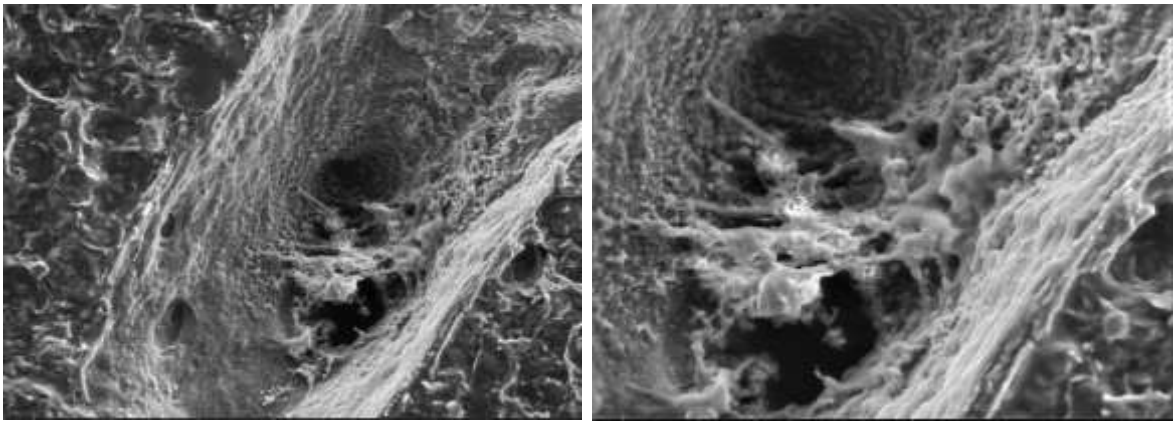
When they study the correlation of macronutrients in the brain layer of the patients with kidney pathology, we found that in comparison with the control group, the oxygen content significantly decreases during stage 1 kidney cancer - by 25% among middle-aged patients, and by 23.8% among elderly patients, 3 stage - by 15% and 15.7%, 4 stage - by 14.7% and 15.3%, respectively (Table 4).

**Table 4: The ratio of macronutrients in patients with kidney pathology (cortical layer)**

| Ratio of macronutrients (%) | | | O | N | C | Ca | S |
|---|---|---|---|---|---|---|---|
| Control | age | Middle (n=10) | 21.25±1.78 | 9.71±1.22 | 67.97±2.13 | 0.14±0.01 | 0.42±0.04 |
| | | Elderly (n=10) | 20.21±1.87 | 9.91±1.31 | 68.98±2.14 | 0.12±0.02 | 0.45±0.04 |
| Kidney cysts | age | Middle (n=10) | 20.14±2.03 | 8.43±1.21 | 70.28±2.23 | 0.14±0.05 | 0.42±0.03 |
| | | Elderly (n=12) | 19.25±1,39 | 8.59±1.18 | 71.12±2.51 | 0.14±0.03 | 0.40±0.02 |
| Kidney cancer stage 1 ($T_1$ $N_0$ $M_0$) | age | Middle (n=10) | 15.83±2.00 | 13.37±1.92 | 69.37±2.31 | 0.38±0.02 | 0.39±0.03 |
| | | Elderly (n=10) | 15.40±1.52* | 12.49±1.31 | 70.76±2.29 | 0.40±0.03 | 0.40±0.04 |
| Kidney cancer stage 2 ($T_1$-$T_2$ $N_0$ $M_0$) | age | Middle (n=12) | 22.91±1.21 | 9.68±1.42 | 63.52±1.39 | 1.15±0.05 | 1.10±0.05* |
| | | Elderly (n=18) | 21.81±1.61 | 9.95±1.29 | 64.42±1.42 | 1.25±0.03 | 1.00±0.03* |
| Kidney cancer stage 3 ($T_1$-$T_2$-$T_3$ $N_1$- $N_2$ $M_0$) | age | Middle (n=13) | 18.05±1.22* | 9.38±1.38 | 73.05±3.21 | 0.62±0.05 | 0.60±0.03* |
| | | Elderly (n=16) | 17.04±1.25* | 9.75±1.65 | 74.15±2.09 | 0.52±0.03 | 0.62±0.04* |
| Kidney cancer stage 4 ($T_1$-$T_2$-$T_3$ $N_1$- $N_2$ $M_1$) | age | Middle (n=8) | 18.11±1.37* | 10.16±1.2 | 69.67±2.38 | 0.51±0.05 | 0.74±0.06* |
| | | Elderly (n=5) | 17.10±1.3* | 11.07±1.33 | 70.81±3.31 | 0.54±0.03 | 0.74±0.07* |

*p<0.05 with respect to the control group

The nitrogen content among the patients with renal pathology did not change significantly as compared with the control group, but there was the tendency to carbon, calcium and sulfur increase.



A B

**Figure 2: A fragment of the renal medulla in organ cancer. Stage 3 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_0$). Man, 64 years old.**

In the area of oncological damage, there are tumor cells near the tubules and blood vessels and inside them located by clones that are loosely connected to each other. The formation of a tumor embolus inside the vessel.

Fig. B (x1000) fragment of Fig. A (x500). SEM.

When they studied the ratio of macronutrients in the cortical layer of the patients with kidney pathology, we found that in comparison with the control group, the oxygen content significantly decreases during stage 1 kidney cancer among middle-aged patients (by 16%), by 12.7% among elderly patients, and it increases during stage 3, and 4 (Table 5)

**Table 5: The ratio of macronutrients in patients with kidney pathology (medulla)**

| Ratio of macronutrients (%) | | | O | N | C | Ca | S |
|---|---|---|---|---|---|---|---|
| Control | age | Middle (n=10) | 21.25±1.78 | 9.71±1.22 | 67.97±2.13 | 0.14±0.01 | 0.42±0.04 |
| | | Elderly (n=10) | 20.21±1.87 | 9.91±1.31 | 68.98±2.14 | 0.12±0.02 | 0.45±0.04 |
| Kidney cysts | age | Middle (n=10) | 18.80±1.99 | 12.09±1.45 | 67.76±2.32 | 0.25±0.04 | 0.45±0.02 |
| | | Elderly (n=12) | 17.84±1.25 | 13.41±1.32 | 66.52±3.36 | 0.25±0.03 | 0.42±0.03 |
| Kidney cancer stage 1 ($T_1$ $N_0$ $M_0$) | age | Middle (n=10) | 17.82±1.56* | 14.41±1.24 | 66.19±3.12 | 0.19±0.03 | 0.39±0.04 |
| | | Elderly (n=10) | 17.63±1.31* | 13.32±1.43 | 67.55±2.46 | 0.21±0.03 | 0.40±0.02 |
| Kidney cancer stage 2 ($T_1$-$T_2$ $N_0$ $M_0$) | age | Middle (n=12) | 20.73±1.24* | 5.93±1.12 | 70.98±3.41 | 0 | 0.37±0.03 |
| | | Elderly (n=18) | 20.32±1.32* | 5.94±1.01 | 71.40±3.12 | 0 | 0.37±0.03 |
| Kidney cancer stage 3 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_0$) | age | Middle (n=13) | 24.14±2.01* | 4.49±1.14 | 69.71±2.87 | 0 | 0.07±0.03* |
| | | Elderly (n=16) | 24.01±1.03* | 4.53±0.45 | 70.00±3.31 | 0 | 0.04±0.01* |
| Kidney cancer stage 4 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_1$) | age | Middle (n=8) | 23.14±2.05* | 5.76±1.23 | 70.20±2.32 | 1.17±0.04 | 1.30±0.14* |
| | | Elderly (n=5) | 22.30±2.03* | 5.81±0.98 | 71.30±2.98 | 1.15±0.03 | 1.10±0.14* |

*$p<0.05$ with respect to the control group

The nitrogen content increases with kidney cysts and during stage 1 kidney cancer, and during stage 2 and 4 it decreases 1.6-1.7 times among middle-aged and elderly patients, during stage 3 - 2.1 times in both age groups. There was a tendency toward carbon increase in some groups of patients, as the content of calcium (8.3 and 9.5 times) and sulfur (3 and 2.4 times) increase in the group of patients with stage 4 kidney cancer.

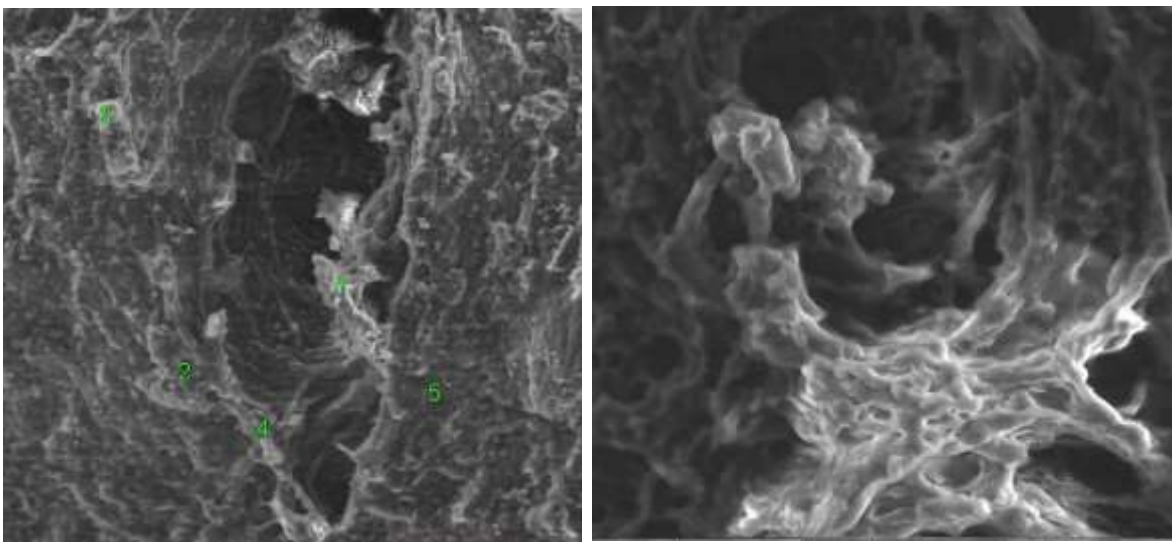
We found that the oxygen content, in comparison with the control group, in case of bladder cancer of stage 3-4 does not significantly decrease, and it decreases by 25.6% for stage 1 and by 49.5% for stage 2 (Table 6).

**Table 6: The ratio of macronutrients in patients with bladder cancer**

| Ratio of macronutrients (%) | | | O | N | C | Ca | S |
|---|---|---|---|---|---|---|---|
| Control | age | Middle (n=10) | 21.25±1.78 | 9.71±1.22 | 67.97±2.13 | 0.14±0.01 | 0.42±0.04 |
| | | Elderly (n=10) | 20.21±1.87 | 9.91±1.31 | 68.98±2.14 | 0.12±0.02 | 0.45±0.04 |
| Bladder cancer stage 1 ($T_1$ $N_0$ $M_0$) | age | Elderly (n=5) | 15.03±1.41* | 13.57±1.68 | 65.78±1.33 | 1.75±0.02 | 1.62±0.04* |
| Bladder cancer stage 2 ($T_1$-$T_2$ $N_0$ $M_0$) | age | Elderly (n=15) | 10.02±1.08* | 13.00±1.07 | 69.89±2.05 | 1.75±0.04 | 1.62±0.08* |
| Bladder cancer stage 3 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_0$) | age | Elderly (n=6) | 19.66±1.71 | 11.19±1.51 | 66.80±2.07 | 0.24±0.03 | 0.69±0.04* |
| Bladder cancer stage 4 ($T_1$-$T_2$-$T_3$ $N_1$-$N_2$ $M_1$) | age | Elderly (n=4) | 19.82±1.94 | 11.22±1.46 | 66.62±3.04 | 0.14±0.02 | 0.69±0.06* |

*p<0.05 with respect to the control group

With this pathology, the tendency towards an insignificant increase of nitrogen and carbon content was noted in all groups, while in the groups of patients with stage 1 and 2 of the bladder cancer, the content of calcium and sulfur increased by 12.5 and 3.8 times, respectively.



A B

**Figure 3: A fragment of the bladder in organ cancer.Stage 3 ($T_1$-$T_2$-$T_3$ $N_1N_2$ $M_0$). Man, 69 years old. Clones of tumor cells inside the vessel (1, 2, 4) with the formation of a tumor embolus (1) and beyond it (3). 1-5 (A) places of definition of elements. Fig. B (x2000) fragment of Fig. A (x500). SEM.**

## SUMMARY

Thus, when they studied macronutrients in oncourological pathology, we found that all groups showed oxygen content decrease, most pronounced among the patients with stage 2 bladder cancer - 49.5%, which leads to tissue hypoxia in the studied organs.

The nitrogen and carbon content varied slightly. Among the patients of all studied groups, the calcium content increases, so: stage 1 prostate cancer - 10.6-10.8 time increase, kidney

cancer - 8.3-9.5 time increase, and bladder cancer - 12.5 time increase. Also, the sulfur content among the patients with kidneys and bladder cancer increased by 2.4 and 3.8 times, respectively.

## CONFLICT OF INTEREST

None

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