Abstract

The aim of the study was to evaluate whether significant changes in the prostatic tissue levels of ratios Zn/trace element contents exist in the malignantly transformed prostate. Contents of 43 trace elements in normal (N, n=37), benign hypertrophic (BPH, n=32) and cancerous human prostate (PCa, n=60) were investigated, and ratios Zn/trace element contents were calculated. Measurements were performed using a combination of non-destructive and destructive methods: instrumental neutron activation analysis and inductively coupled plasma mass spectrometry, respectively. It was observed that the Zn/Ag, Zn/Al, Zn/Au, Zn/Bi, Zn/Be, Zn/Bi, Zn/Br, Zn/Cd, Zn/Ce, Zn/Co, Zn/Cr, Zn/Cs, Zn/Dy, Zn/Er, Zn/Fe, Zn/Gd, Zn/Hg, Zn/Ho, Zn/In, Zn/Li, Zn/Mn, Zn/Mo, Zn/Nb, Zn/Nd, Zn/Ne, Zn/Pb, Zn/Pt, Zn/Rh, Zn/Sb, Zn/Se, Zn/Sm, Zn/Sn, Zn/Tb, Zn/Tl, Zn/Ti, Zn/Tm, Zn/U, Zn/Y, Zn/Yb, and Zn/Zr mass fraction ratios were significantly lower in cancerous tissue than in normal and BPH prostate. Finally, we propose to use the Zn/Ag, Zn/Al, Zn/Au, Zn/Bi, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rh, and Zn/Sb mass fraction ratios in a needle biopsy core as an accurate tool to diagnose prostate cancer. Further studies on larger number of samples are required to confirm our findings and to investigate the impact of the trace element relationships on prostate cancer etiology.

Introduction

The prostate gland may be a source of many health problems in men past middle age, the most common being benign prostatic hyperplasia (BPH), and prostate carcinoma (PCa). BPH is a noncancerous enlargement of the prostate gland leading to obstruction of the urethra and can significantly impair quality of life. The prevalence of histological BPH is found in approximately 50-60% of males age 40-50, in over 70% at 60 years old and in greater than 90% of men over 70 [1,2]. In many Western industrialized countries, including North America, PCa is the most frequently diagnosed form of noncutaneous malignancy in males and, except for lung cancer, is the leading cause of death from cancer [3-8]. Although the etiology of BPH and PCa is unknown, some trace elements have been highlighted in the literature in relation to the development of these prostate diseases [9-29].

Trace elements have essential physiological functions such as maintenance and regulation of cell function and signalling, gene regulation, activation or inhibition of enzymatic reactions, neurotransmission, and regulation of membrane function. Essential or toxic (mutagenic, carcinogenic) properties of trace elements depend on tissue-specific need or tolerance, respectively [30]. Excessive accumulation, deficiency or an imbalance of the trace elements may disturb the cell functions and may result in cellular degeneration, death and malignant transformation [30].

In reported studies significant changes of trace element contents in hyperplastic and cancerous prostate in comparison with those in the normal prostatic tissue were observed [31-64]. Moreover, a significant informative value of Zn content as a tumor marker for PCa diagnostics was shown by us [65,66]. Hence its possible that besides Zn, some other trace elements also can be used as tumor markers for distinguish between benign and malignant prostate.

Current methods applied for measurement of trace elements contents in samples of human tissue include a number of methods. Among thesemethods the instrumental neutron activation analysis with high resolution spectrometry of long-lived radionuclides (INAA-LLR) is a non-destructive and one of the most sensitive techniques. It allows measure the trace element contents in a few milligrams tissue without any treatment of sample. Analytical studies of the Ag, Co, Cr, Fe, Hg, Hg, Pb, Sc, Se, and Zn contents in normal, BPH and PCa tissue were done by us using INAA-LLR [15,20,28,60,62]. Nondestructive method of analysis avoids the possibility of changing the content of trace elements in the studied samples [67-70], which allowed for the first time to obtain reliable results. In particular, it was shown that the average mass fraction of Co, Cr, Hg, Hg, and Se in BPH were higher than normal levels [66], but in PCa tissues the mean values of Ag, Cr, Fe, Hg, Hg, and Sb were lower while those of Co, Rb, Sc, and Zn were lower than in healthy prostatic tissue [60,66]. Obtained results formed the basis for a new method for differential diagnosis of BPH and PCa, the essence of which was to determine the ratios of trace element contents changedin opposite directions during malignant transformation of prostate.

It is obvious that the most effective will be non-destructive
analytical methods because they involve a minimal treatment of sample since the chances of significant loss or contamination would be decreased. However, the INAA-LLR allow only determine the mean mass fractions of 9-10 trace elements in the samples of normal and cancerous prostate glands [15,20,28,60,66]. The inductively coupled plasma mass spectrometry (ICP-MS) is a more power analytical tool than INAA-LLR [18] but sample digestion is a critical step in elemental analysis by this method. In the present study both analytical methods were used and the results obtained for some trace elements by ICP-MS were under the control of INAA-LLR data.

The present study had three aims. The main objective was to obtain reliable results about the 43 trace elements: Ag, Al, Au, B, Be, Bi, Br, Cd, Ce, Co, Cr, Cs, Dy, Er, Fe, Gd, Hg, Ho, La, Li, Mn, Mo, Nb, Nd, Ni, Pb, Pr, Rh, Sc, Se, Sm, Sn, Tb, Th, Ti, Tl, Tm, U, Y, Yb, Zn, and Zr contents in intact prostate of healthy men aged over 40 years and in the prostate gland of age-matched patients, who had either BPH or PCA combining in consecutive order non-destructive INAA-LLR with destructive ICP-MS. The second aim was to calculate Zn/trace element content ratios and compare the levels of these ratios in normal, hyperplastic, and cancerous prostate. The third and final aim was to evaluate the ratios of Zn/trace element contents for diagnosis of prostate cancer.

All studies were approved by the Ethical Committees of the Medical Radiological Research Centre, Obninsk.

Material and methods

Samples

The patients studied (n=92) were hospitalized in the Urological Department of the Medical Radiological Research Centre. All of them were European-Caucasian, citizens of Moscow and Obninsk (a small city in a non-industrial region 105 km south-west of Moscow). Transrectal puncture biopsy of suspicious indurated regions of the prostate was performed for every patient, to permit morphological study of prostatic tissue at these sites and to estimate their chemical element contents. In all cases the diagnosis has been confirmed by clinical and morphological results obtained during studies of biopsy and resected materials. The age of 32 patients with BPH ranged from 56 to 78 years, the mean being 66±6 (M±SD) years. The 60 patients aged 40-79 suffered from PCA (stage T1-T4). Their mean age was 65±10 (M±SD) years.

Intact prostates (N) were removed at necropsy from 37 men aged 41-87 who had died suddenly. All deceased were European-Caucasian, citizens of Moscow. Their mean age was 551±11 (M±SD) years. The majority of deaths were due to trauma. Tissue samples were collected from the peripheral zone of dorsal and lateral lobes of their prostates, within 2 days of death and then the samples were divided into two portions. One was used for morphological study while the other was intended for trace element analysis. A histological examination was used to control the age norm conformity, as well as to confirm the absence of microadenomatosis and latent cancer [15,20,28].

Sample preparation

All tissue samples were divided into two portions. One was used for morphological study while the other was intended for trace element analysis. After the samples intended for trace element analysis were weighed, they were freeze-dried and homogenized. The sample weighing about 10 mg (for biopsy materials) and 50-100 mg (for resected materials) was used for trace element measurement by INAA-LLR. The samples for INAA-LLR were wrapped separately in a high-purity aluminum foil washed with double rectified alcohol beforehand and placed in a nitric acid-washed quartz ampoule.

After NAA-LLR investigation the prostate samples were taken out and used for ICP-MS. The samples were decomposed in autoclaves; 1.5 mL of concentrated HNO3 (nitric acid at 65%, maximum (max) of 0.0000005% Hg; GR, ISO, Merck) and 0.3 mL of H2O2 (pure for analysis) were added to prostate tissue samples, placed in one-chamber autoclaves (Ancon-AT2, Ltd., Russia) and then heated for 3 h at 160–200°C. After autoclaving, they were cooled to room temperature and solutions from the decomposed samples were diluted with deionized water (up to 20 mL) and transferred to plastic measuring bottles. Simultaneously, the same procedure was performed in autoclaves without tissue samples (only HNO3+H2O2+deionized water), and the resultant solutions were used as control samples.

Instrumentation and methods

A vertical channel of a nuclear reactor was applied to determine the trace element mass fractions by NAA-LLR. The quartz ampoule with prostate samples and certified reference materials was soldered, positioned in a transport aluminum container and exposed to a 24-hour neutron irradiation in a vertical channel with a neutron flux of 1.3⋅109 n cm−2 s−1. Ten days after irradiation samples were reweighed and repacked. The samples were measured for period from 10 to 30 days after irradiation. The duration of measurements was from 20 min to 10 hours subject to pulse counting rate. The gamma spectrometer included the 100 cm² Ge(Li) detector and on-line computer-based multichannel analyzer. The spectrometer provided a resolution of 1.9 keV on the 60Co 1332 keV line.

An ICP-MS Thermo-Fisher “X-7” Spectrometer (Thermo Electron, USA) was used to determine the content of trace elements by ICP-MS. The element concentrations in aqueous solutions were determined by the quantitative method using multi elemental calibration solutions ICP-MS-68A and ICP-AM-6-А produced by High-Purity Standards (Charleston, SC 29423, USA). Indium was used as an internal standard in all measurements.

Information detailing with the NAA-LLR and ICP-MS methods used and other details of the analysis was presented in our previous publication [15,20,28,60,66].

Certified reference materials

For quality control, ten subsamples of the certified reference materials IAEA H-4 Animal muscle from the International Atomic Energy Agency (IAEA), and also five sub-samples INCT-SBF-4 Soya Bean Flour, INCT-TL-1 Tea Leaves and INCT-MPH-2 Mixed Polish Herbs from the Institute of Nuclear Chemistry and Technology (INCT, Warszawa, Poland) were analyzed simultaneously with the investigated prostate tissue samples. All samples of CRM were treated in the same way as the prostate samples. Detailed results of this quality assurance program were presented in earlier publications [15,18,22].

Computer programs and statistics

A dedicated computer program for INAA mode optimization was used [21]. All prostate samples for INAA-LLR were prepared in duplicate and mean values of chemical element contents were used in final calculation. For elements investigated by INAA-LLR and ICP-MS the mean of all results was used. Using the Microsoft Office Excel software Zn/trace element contents for each trace element in every
sample were calculated. Then arithmetic mean, standard deviation, and standard error of mean were calculated for ratios of Zn/trace element mass fraction in normal, benign hyperplastic and cancerous prostate tissue. The difference in the results between BPH and Norm, Pca and Norm, and PCA and BPH was evaluated by parametric Student’s t-test and non-parametric Wilcoxon–Mann–Whitney U-test. Values of p<0.05 were considered to be statistically significant. For the construction of "individual data sets for Zn/trace element mass fraction ratios in normal, benign hypertrophic and cancerous prostate" diagrams the Microsoft Office Excel software was also used.

Results

Table 1. Comparison of mean values (M ± SEM) of the Zn mass fraction/trace element mass fraction ratios in normal (N), benign hyperplastic (BPH) and cancerous prostate (PCa).

| Element | Prostatic tissue | Normal 41-87 year (n=37) | BPH 56-78 year (n=32) | PCa 40-79 year (n=60) |
|---------|------------------|---------------------------|----------------------|----------------------|
| Zn/Ag   |                  | 37079 ± 5748              | 53275 ± 10549        | 1441 ± 364           |
| Zn/Al   |                  | 39.5 ± 8.3                | 58.9 ± 9.6           | 3.34 ± 0.56          |
| Zn/Au   |                  | 597386 ± 199885           | 789472 ± 148247      | 18620 ± 10630        |
| Zn/B     |                  | 1864 ± 472                | 1276 ± 324           | 33.4 ± 22.7          |
| Zn/Be   |                  | 1166728 ± 171086          | 1569218 ± 246879     | 26804 ± 15433        |
| Zn/Bi   |                  | 222186 ± 5247             | 75425 ± 36054        | 2582 ± 2292          |
| Zn/Bz   |                  | 51.6 ± 9.5                | 69.1 ± 11.4          | 2.39 ± 0.84          |
| Zn/Cd   |                  | 1451 ± 356                | 2488 ± 533           | 382 ± 44             |
| Zn/Ce   |                  | 58752 ± 13885             | 126472 ± 18956       | 2207 ± 812           |
| Zn/Cr   |                  | 5303 ± 1858               | 1952 ± 687           | 116 ± 34             |
| Zn/Cs   |                  | 36214 ± 7979              | 65687 ± 12084        | 4989 ± 1601          |
| Zn/Dy   |                  | 641749 ± 143308           | 1125693 ± 231414     | 3543 ± 15486         |
| Zn/Er   |                  | 114919 ± 258895           | 2659891 ± 510876     | 66123 ± 18514        |
| Zn/Fa   |                  | 11.3 ± 1.5                | 11.8 ± 1.3           | 1.04 ± 0.11          |
| Zn/Gd   |                  | 569553 ± 138780           | 113840 ± 205832      | 27895 ± 9282         |
| Zn/Hg   |                  | 30732 ± 5037              | 5720 ± 582           | 1553 ± 314           |
| Zn/Ho   |                  | 3132261 ± 701937          | 5325633 ± 85569      | 108255 ± 37392       |
| Zn/La   |                  | 57719 ± 20755             | 91631 ± 18966        | 2156 ± 1195          |
| Zn/Li   |                  | 33798 ± 7925              | 48938 ± 9863         | 1442 ± 559           |
| Zn/Mn   |                  | 814 ± 17                  | 1222 ± 167           | 45.1 ± 19.9          |
| Zn/Mo   |                  | 4789 ± 827                | 8786 ± 1500          | 727 ± 325            |
| Zn/Nb   |                  | 556870 ± 200334           | 681455 ± 152014      | 56921 ± 14279        |
| Zn/Nd   |                  | 129475 ± 31107            | 260623 ± 39406       | 5274 ± 1715          |
| Zn/Ni   |                  | 669 ± 171                 | 770 ± 198            | 30.1 ± 11.2          |
| Zn/Pb   |                  | 1478 ± 339                | 2885 ± 497           | 139 ± 60             |
| Zn/Pr   |                  | 517616 ± 132976           | 1395347 ± 305643     | 26153 ± 8906         |
| Zn/Rb   |                  | 86.9 ± 11.8               | 95.8 ± 10.7          | 18.8 ± 2.1           |
| Zn/Sb   |                  | 45850 ± 10375             | 21798 ± 6086         | 702 ± 250            |
| Zn/Sc   |                  | 58691 ± 12144             | 60497 ± 15015        | 23410 ± 8832         |
| Zn/Se   |                  | 1495 ± 176                | 1449 ± 345           | 296 ± 36             |
| Zn/Sn   |                  | 617945 ± 137213           | 1726655 ± 386992     | 32773 ± 10145        |
| Zn/Sr   |                  | 7831 ± 1944               | 19576 ± 3420         | 349 ± 176            |
| Zn/Tb   |                  | 554412 ± 138287           | 10103881 ± 1779214   | 250461 ± 96820       |
| Zn/Tc   |                  | 699184 ± 187605           | 991175 ± 183154      | 11971 ± 6518         |
| Zn/Tl   |                  | 1014 ± 104                | 1054 ± 150           | 42.0 ± 2.1           |
| Zn/Tm   |                  | 828929 ± 108263           | 879374 ± 135511      | 33512 ± 22230        |
| Zn/Tn   |                  | 748440 ± 2080528          | 1145820 ± 1965135    | 475767 ± 262385      |
| Zn/Tp   |                  | 1454390 ± 90481           | 1437144 ± 312075     | 29068 ± 6153         |
| Zn/Tq   |                  | 169174 ± 5279             | 256865 ± 46436       | 5305 ± 1507          |
| Zn/Tb   |                  | 1503735 ± 356906          | 3084223 ± 755203     | 100641 ± 29271       |
| Zn/Tc   |                  | 48858 ± 9972              | 42437 ± 13129        | 181 ± 59             |

M: Arithmetic mean; SEM: Standard error of mean; NS: not significant difference.

*Titanium tools were used for sampling and preparation.

The ratios of means and the difference between mean values of the Zn/Ag, Zn/Al, Zn/Au, Zn/B, Zn/Be, Zn/Bi, Zn/Br, Zn/Cd, Zn/Ce, Zn/Co, Zn/Cr, Zn/Cs, Zn/Cd, Zn/Dy, Zn/Er, Zn/Fe, Zn/Gd, Zn/Hg, Zn/Ho, Zn/La, Zn/Li, Zn/Mn, Zn/Mo, Zn/Nb, Zn/Nd, Zn/Ni, Zn/Pb, Zn/Pr, Zn/Rb, Zn/Sb, Zn/Sc, Zn/Se, Zn/Sm, Zn/Sn, Zn/Tb, Zn/Th, Zn/Ti, Zn/Tl, Zn/Tm, Zn/U, Zn/Y, Zn/Yb, and Zn/Zr mass fraction ratios in normal, benign hypertrophic and cancerous prostate were presented in Table 2.

Table 3 contains parameters of the importance (sensitivity, specificity and accuracy) of Zn/Ag, Zn/Al, Zn/B, Zn/Be, Zn/Bi, Zn/Br, Zn/Cd, Zn/Ce, Zn/Co, Zn/Cr, Zn/Cs, Zn/Dy, Zn/Er, Zn/Fe, Zn/Gd, Zn/Hg, Zn/Ho, Zn/La, Zn/Li, Zn/Mn, Zn/Mo, Zn/Nb, Zn/Nd, Zn/Ni, Zn/Pb, Zn/Pr, Zn/Rb, Zn/Sb, Zn/Sc, Zn/Se, Zn/Sm, Zn/Sn, Zn/Tb, Zn/Th, Zn/Ti, Zn/Tl, Zn/Tm, Zn/U, Zn/Y, Zn/Yb, and Zn/Zr mass fraction ratios in normal, benign hypertrophic and cancerous prostate, respectively.
Table 2. Ratio of means and the difference between mean values of the Zn mass fraction/trace element mass fraction ratios in normal (N), benign hypertrophic (BPH) and cancerous prostate (PCa).

| Element Pair | BPH and Normal (N) | PCa and Normal (N) | PCa and BPH |
|--------------|--------------------|--------------------|--------------|
| Zn/Ag        | 1.44               | 0.187              | >0.05        |
| Zn/Al        | 1.49               | 0.140              | >0.05        |
| Zn/Au        | 1.32               | 0.445              | >0.05        |
| Zn/B         | 0.68               | 0.312              | >0.05        |
| Zn/Be        | 1.34               | 0.195              | >0.05        |
| Zn/Cd        | 0.34               | 0.027              | ≤0.05        |
| Zn/Ce        | 2.15               | 0.0089             | ≤0.05        |
| Zn/Co        | 0.78               | 0.291              | >0.05        |
| Zn/Cr        | 0.39               | 0.129              | >0.05        |
| Zn/Cs        | 1.81               | 0.056              | ≤0.05        |
| Zn/Cy        | 1.75               | 0.072              | ≤0.05        |
| Zn/Er        | 2.31               | 0.018              | ≤0.05        |
| Zn/Fe        | 1.04               | 0.814              | >0.05        |
| Zn/Gd        | 1.91               | 0.042              | ≤0.05        |
| Zn/Hg        | 0.19               | 0.000026           | ≤0.01        |
| Zn/Ho        | 1.70               | 0.059              | ≤0.05        |
| Zn/La        | 1.59               | 0.238              | >0.05        |
| Zn/Li        | 1.45               | 0.244              | >0.05        |
| Zn/Mn        | 1.50               | 0.102              | >0.05        |
| Zn/Mo        | 1.83               | 0.033              | ≤0.05        |
| Zn/Nb        | 1.22               | 0.624              | >0.05        |
| Zn/Nd        | 2.05               | 0.012              | ≤0.05        |
| Zn/Ni        | 1.12               | 0.757              | >0.05        |
| Zn/Pb        | 1.95               | 0.030              | ≤0.05        |
| Zn/Pr        | 2.70               | 0.036              | ≤0.05        |
| Zn/Rh        | 1.10               | 0.578              | >0.05        |
| Zn/Sb        | 0.48               | 0.051              | ≤0.05        |
| Zn/Sc        | 1.03               | 0.926              | >0.05        |
| Zn/Sn        | 0.97               | 0.906              | >0.05        |
| Zn/Sm        | 2.79               | 0.019              | ≤0.05        |
| Zn/Su        | 2.50               | 0.0084             | ≤0.05        |
| Zn/Tb        | 1.82               | 0.055              | ≤0.05        |
| Zn/Th        | 1.42               | 0.275              | >0.05        |
| Zn/Ti        | 1.50               | 0.098              | ≤0.05        |
| Zn/Tl        | 1.06               | 0.774              | >0.05        |
| Zn/Tm        | 1.33               | 0.176              | >0.05        |
| Zn/U         | 3.16               | 0.011              | ≤0.05        |
| Zn/V         | 1.52               | 0.221              | ≥0.05        |
| Zn/Wb        | 2.05               | 0.079              | >0.05        |
| Zn/Xr        | 0.87               | 0.711              | >0.05        |

p≤ - Student’s t-test, U-test - Wilcoxon-Mann-Whitney U-test, Bold significant differences

Analysis the mass fraction ratios of trace element in prostate tissue could become a powerful diagnostic tool. To a large extent, the resumption of the search for new methods for early diagnosis of PCa was due to experience gained in a critical assessment of the limited capacity of the prostate specific antigen (PSA) serum test [72]. In addition to the PSA serum test and morphological study of needle-biopsy cores of the prostate, the development of other highly precise testing methods seems to be very useful. Experimental conditions of the present study were approximated to the hospital conditions as closely as possible. In BPH and PCa cases we analyzed a part of the material obtained from a puncture transrectal biopsy of the indurated site in the prostate. Therefore, our data allow us to evaluate adequately the importance of trace element mass fraction ratios for the diagnosis of PCa. As is evident from Table 2 and, particularly, from individual data sets (Figure 1), the Zn/Ag, Zn/Al, Zn/B, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rh, and Zn/Sb mass fraction ratios are potentially the most informative test for a differential diagnosis. For example, if 5.0 is the value of Zn/Al mass fraction ratio assumed to be the upper limit for PCa (Figure 1) and an estimation is made for “PCa or intact and BPH tissue”, the following values are obtained:

Sensitivity=[True Positives (TP)/[TP+False Negatives (FN)]]×100%=100-9%;
Specificity=[True Negatives (TN)/[TN+False Positives (FP)]]×100%=97±3%.
Figure 1. Individual data sets for Zn/Ag, Zn/Al, Zn/B, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rb, and Zn/Sb mass fraction ratios in samples of normal (1), benign hypertrophic (2) and cancerous (3) prostate

Table 3. Parameters of the importance (sensitivity, specificity and accuracy) of Zn/Ag, Zn/Al, Zn/B, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rb, and Zn/Sb mass fraction ratios for the diagnosis of PCa (an estimation is made for “PCa or normal and BPH prostate”).

| Mass fraction ratio or their multiplication | Upper limit for PCa | Sensitivity % | Specificity % | Accuracy % |
|-------------------------------------------|---------------------|---------------|---------------|------------|
| Zn/Ag                                     | 4500                | 97 ± 3        | 100-2         | 99 ± 1     |
| Zn/Al                                     | 5.0                 | 100-9         | 97 ± 3        | 98 ± 2     |
| Zn/B                                      | 100                 | 90 ± 10       | 100-3         | 98 ± 2     |
| Zn/Be                                     | 180000              | 100-9         | 98 ± 2        | 96 ± 3     |
| Zn/Br                                     | 3.5                 | 91 ± 6        | 100-2         | 97 ± 2     |
| Zn/Cr                                     | 260                 | 93 ± 5        | 98 ± 2        | 96 ± 2     |
| Zn/Fe                                     | 2.0                 | 87 ± 5        | 98 ± 2        | 94 ± 3     |
| Zn/Mn                                     | 110                 | 91 ± 9        | 100-3         | 98 ± 2     |
| Zn/Rb                                     | 33                  | 92 ± 4        | 89 ± 4        | 91 ± 3     |
| Zn/Sb                                     | 2000                | 95 ± 4        | 96 ± 3        | 96 ± 2     |
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The number of people (samples) examined was taken into account for calculation of confidence intervals [73]. In other words, if Zn/Al mass fraction ratio in a prostate biopsy sample is lower than 0.1, one could diagnose a malignant tumor with an accuracy 98±2%. Thus, using the Zn/Al mass fraction ratio-test makes it possible to diagnose cancer in 100%-9% cases (sensitivity). The same way parameters of the importance (sensitivity, specificity and accuracy) of Zn/Ag, Zn/B, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rb, and Zn/Sb mass fraction ratios for the diagnosis of PCa were calculated (Table 3).

Conclusion

The combination of nondestructive INAA-ALSLR and destructive ICP-MS methods is satisfactory analytical tool for the precise determination of 43 trace element mass fractions in the tissue samples of normal, BPH and carcinomatous prostate glands. The sequential application of two methods allowed precise quantitative determinations of mean mass fraction of Ag, Al, Au, B, Be, Bi, Br, Cd, Ce, Co, Cr, Cs, Dy, Er, Fe, Gd, Hg, Ho, La, Li, Mn, Mo, Nb, Nd, Ni, Pb, Pr, Rh, Sb, Sc, Se, Sm, Sn, Tb, Th, Ti, Tl, Tm, U, Y, Yb, Zn and Zr. It was observed that the Zn/Ag, Zn/Al, Zn/Au, Zn/B, Zn/Be, Zn/Br, Zn/Cd, Zn/Ce, Zn/Cr, Zn/Cu, Zn/Dy, Zn/Er, Zn/Fe, Zn/Gd, Zn/Hg, Zn/Ho, Zn/La, Zn/Li, Zn/Mn, Zn/Mo, Zn/Nb, Zn/Nd, Zn/Ni, Zn/Pb, Zn/Pr, Zn/Rb, Zn/Sb, Zn/Sc, Zn/Se, Zn/Sm, Zn/Sn, Zn/Tb, Zn/Th, Zn/Ti, Zn/Tl, Zn/Tm, Zn/U, Zn/Y, Zn/Yb, and Zn/Zr mass fraction ratios were significantly lower in cancerous tissues than in normal and BPH prostate. Finally, we propose to use the Zn/Ag, Zn/Al, Zn/B, Zn/Be, Zn/Br, Zn/Cr, Zn/Fe, Zn/Mn, Zn/Rb, and Zn/Sb mass fraction ratios in a needle-biopsy core as an accurate tool to diagnose prostate cancer. Further studies on larger number of samples are required to confirm our findings and to investigate the impact of the trace element relationships on prostate cancer etiology.

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Competing interests

All other authors declare no competing interests.

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