Comparison of Trace Metal Concentrations in Malign and Benign Human Prostate

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Imbalance in the composition of trace metals, recognized to be essential to normal human homeostasis, besides accumulation of potentially toxic or nonessential trace metals, may cause disease. The essential trace elements have four major functions as stabilizers, elements of structure, essential elements for hormonal function, and cofactors in enzymes. As a result, the lack of essential trace elements will influence structure alone or will alter function of structure through the lack of stabilization, change of charge properties, or allosteric configuration. It may be expected that the deficiency of essential trace elements as cofactors of enzymes could severely impair the host's resistance against carcinogenic stress. From these elements, zinc is a component of over 300 proteins and over 100 DNA-binding proteins with zinc fingers. Zn and Cu are the prosthetic groups of some metalloenzymes containing superoxide dismutase (SOD), which is an important antioxidant enzyme for cellular protection from reactive oxygen species (ROS). It is described that benign prostatic obstruction (BPO) is characterized by high Zn concentrations and prostate cancer (PCa) is characterized by low Zn concentrations.

Cancer is a multifactorial and multifactorial complex disease. The role of metals in the development and inhibition of cancer has a complex character and raises many questions. In the last 20 years, some metals, including cadmium, nickel, arsenic, cobalt, and chromium(VI), were recognized as human or animal carcinogens in addition to primary carcinogens such as radiation, viruses, and other chemicals. Their carcinogenic potentials depend largely on factors such as oxidation states and solubilities. The induction of oxidative DNA damage and the interaction with DNA repair processes were leading to an enhancement of genotoxicity in combination with a variety of DNA-damaging agents. Nucleotide excision repair (NER), which is the major repair system, is inhibited at low levels as well as at nontoxic concentrations of NiII, CdII, CoII, and AsIII. The repair of oxidative DNA base modifications is disturbed by NiII and CdII. One reason for repair inhibition appears to be the displacement of ZnII and MgII. Mg and Zn, essential elements that are cofactors for DNA polymerase, are effective protectors against carcinogenesis in vivo.

The most common analytical technique used for trace metal analysis in biological matrices is atomic absorption spectrometry. For improved sensitivity of flame atomic absorption spectrometry (FAAS), a slotted atom trap (STAT) was used for some metals such as Cd and Pb. In this study, the concentrations of different trace metals, including Cd, Ni, Cu, Zn, Fe, Mg, and Ca in malign and benign prostate tissues, were determined by atomic absorption spectrophotometry. For digestion of the tissues, a microwave oven was used.

Results and Discussion

Calibration curves were obtained by using the solutions of the studied elements at different concentrations.
were found to be the same as those obtained with the additions method. The slopes of the calibration curves for all studied elements were possible interferences caused by the matrix. The slopes of the calibration curves were parallel to the calibration curves and the recoveries were found for Ca (0.25–3.0 mg/L) by STAT-AAS.

The graphs obtained were linear in the concentration range described follow, and the equations of the curves were as follows:

\[ Y = 1.11X + 0.4 \quad R^2 = 0.99 \]

for Cd (10–140 ng/mL by STAT-AAS)

\[ Y = 85X + 0.5 \quad R^2 = 0.99 \]

for Ni (0.25–2.0 mg/L)

\[ Y = 0.21X + 0.5 \quad R^2 = 1.0 \]

for Cu (50–500 ng/mL by STAT-AAS)

\[ Y = 302X + 0.75 \quad R^2 = 0.99 \]

for Zn (0.1–1.0 mg/L)

\[ Y = 64X +0.43 \quad R^2 = 0.99 \]

for Fe (0.20–3.0 mg/L)

\[ Y = 515X +7.0 \quad R^2 = 0.99 \]

for Mg (0.25–2.0 mg/L)

\[ Y = 305X + 39 \quad R^2 = 0.99 \]

for Ca (0.25–2.0 mg/L).

The accuracy of the method was studied by examining the recovery of the metals from prostate samples fortified with various amounts of the studied metals. The following metal amounts were added: 50 ng/g of Cd, 100 ng/g of Ni, 0.5 mg/kg of Cu, 40 mg/kg of Zn, 10 mg/kg of Fe, 100 mg/kg of Mg, and 200 mg/kg of Ca. After digestion by microwave oven, the recoveries were found to be at least 93% for all studied metals. In addition, the standard additions method was used to investigate possible interferences caused by the matrix. The slopes of the calibration curves for all studied elements were compared with the slopes obtained by the standard additions method. The slopes of the calibration curves were found to be the same as those obtained with the standard additions method. In other words, all of the standard additions curves were parallel to the calibration curves. These results indicate an absence of chemical interference.

**Comparison of Metal Levels in the Malign and Benign Tissues.** Data related with carcinogenic effects of cadmium and nickel have been detailed in the literature. Occupational exposure to Cd is associated with lung cancers in humans, whereas other sites, potentially including the prostate, are not definitively established. Cadmium can also cause prostatic proliferative lesions, including adenocarcinomas after systemic or direct exposure. As seen from Table 2, high Cd concentrations were found in both malignant and benign prostate tissues. The average Ni contents in the studied malign prostate samples were found to be significantly higher (p < 0.1) than those in the benign samples.

**Zinc.** It has been known that Zn concentration in the prostate gland is much higher than in other human tissues. In addition, there is some evidence that an increase of Zn content in benign prostatic hyperplasia (BPH) and a decrease in prostatic carcinoma (PCA), as compared to normal tissues, occurs, but the information about the mechanism of the accumulation of Zn in prostate is too incomplete to draw any conclusions regarding its importance. However, Zn and Se deficiency were considered as possible cancer risk factors for prostate.

In contrast to the above literature information, it is seen from Table 2 and Figure 1 that the Zn levels in the malign prostate tissues were significantly higher than the Zn levels in the benign prostate tissues (p < 0.05). One cause of these results may be Zn accumulated in apoptotic cells because cell homeostasis was destroyed. Reactive oxygen substances (ROS) could produce DNA damage with toxic free hydroxyl radicals in vivo and lead to cancer. The prostate converts testosterone to dihydrotestosterone, a key substrate for downstream hormone metabolism. Withdrawal of testosterone by surgical or medical castration is a well-known treatment for prostate cancer and is effective in

| Table 2. Trace Metal Concentrations in the Malignant and Benign Prostate Tissues |
|-----------------|----------------|----------------|----------------|----------------|
| tissue          | Cd, ng/g       | Ni, ng/g       | Cu, mg/kg      | Zn, mg/kg      |
| prostate        | malignant      | benign         | malignant      | benign         |
| prostate        | 124 ± 13       | 33 ± 4         | 1300 ± 100     | 75 ± 8         |
| prostate        | 100 ± 12       | 130 ± 15       | 200 ± 26       | 500 ± 60       |
| prostate        | 80 ± 9         | 120 ± 12       | 120 ± 13       | 100 ± 9        |
| prostate        | 65 ± 7         | 20 ± 2         | 400 ± 46       | 450 ± 55       |
| prostate        | 62 ± 7         | 74 ± 8         | 500 ± 58       | 400 ± 52       |
| prostate        | 49 ± 5         | 105 ± 11       | 480 ± 50       | 300 ± 50       |
| prostate        | 65 ± 5         | 630 ± 70       | 49 ± 0.1       | 43 ± 5         |
| prostate        | 66 ± 6         | 440 ± 50       | 0.5 ± 0.1      | 47 ± 5         |
| prostate        | 56 ± 5         | 301 ± 30       | 0.7 ± 0.1      | 45 ± 5         |
| prostate        | 90 ± 10        | 85 ± 8         | 500 ± 63       | 350 ± 40       |
| prostate        | 25 ± 3         | 330 ± 32       | 0.2 ± 0.03     | 46 ± 4         |
| prostate        | 25–124         | 20–130         | 130–1300       | 75–500         |
| prostate        | 240–62         | 240–1350       | 750–700        | 1350–2050      |
| prostate        | 11 ± 1         | 13 ± 1         | 1300 ± 650     | 825 ± 35       |
| prostate        | 0.4 ± 0.1      | 0.3 ± 0.1      | 19 ± 1         | 42 ± 6         |
| prostate        | 0.3 ± 0.1      | 0.3 ± 0.1      | 19 ± 1         | 42 ± 6         |
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* The results are mean values (fresh weight basis), n = 3. Every malign and benign tissue belongs to a different person. Standard deviations for the Ca results of prostate are in the range 15–50%. The malign and benign tissues in this line are belong to the same person.
75–80% of patients with metastatic prostate cancer.\textsuperscript{19} Zn can inhibit transforming of testosterone to dihydrotestosterone at low and also at high concentrations too.\textsuperscript{18} Therefore, the second cause of high Zn concentration in the malign prostate may be the result of prostate cancer due to inhibition of the transformation of testosterone to dihydrotestosterone.

The differences between the literature in which Zn levels in malign prostate were lower than those in the benign prostate and our values that show Zn levels in malign prostate were higher than those in the benign prostate may be caused for the following reasons: (a) Zn is not uniformly distributed throughout the different anatomical regions of the prostate. For example, the Zn levels in the lateral lobe and anterior lobe were 211 and 84 mg/kg on a wet tissue basis, respectively.\textsuperscript{15} (b) The literature values were either too old (up to 35 years) \textsuperscript{15} or were given on a dry weight basis.\textsuperscript{16}

**Iron.** Reducer active metal ions, such as Fe and Cu, play a role in the increase of the ROS production (Fenton reaction) in biological systems.\textsuperscript{20} Although Fe is an essential nutritional element for all life forms, it is known that excess iron, like iron deficiency, also leads to oxidative DNA damage.\textsuperscript{21} Similar to zinc, iron levels in the malign prostate tissues were found to be higher ($p < 0.1$) than the levels in the benign prostate tissues (Figure 2).

**Copper.** Although copper is an essential element for humans and animals, high concentrations of Cu (above normal) could induce growth proliferation and cancer by damaging DNA with toxic free hydroxyl radicals.\textsuperscript{20} In this study, Cu levels were found to be slightly higher ($p < 0.2$) in the malign prostate samples than in the benign prostate samples (Table 2 and Figure 3). Unfortunately, we did not measure the ROS levels in the studied prostate tissues. Therefore, there is a need for studies regarding the ROS production and the trace element levels.

**Magnesium and Calcium.** As similar to Zn, Fe, and Cu, the magnesium levels were also found to be higher ($p = 0.1018$) in the malign prostate samples than in the benign prostate samples (Figure 4). Last, calcium levels in the malign prostate samples were found to be 2 times higher ($p < 0.05$) than those in the benign samples (Figure 5). This suggested that the stored calcium in the cells was not departing from the cells because the cell functions were disturbed in the malign cancer.

In Table 2, there are also results belonging the same person for malign and benign samples. It was observed that similar results to the above were found for these samples.
In the Table 3, significant and tendentious elements are listed. The positive sign is used to illustrate the accumulation of the element in malign prostate, and the minus sign is used to indicate the depletion of the element in malign prostate.

In conclusion, we have found that zinc, iron, copper, nickel, magnesium, and calcium concentrations in malignant prostate samples were higher than those in the benign prostate samples, in contrast to the literature data for zinc. We think that the increase in Ca levels and its heterogeneous distribution in malign samples are very important for the investigation of cancer and its heterogeneous distribution in malign samples except one sample. In addition, two urinary bladder samples were taken. The tissue samples were cut into small pieces with a stainless steel knife and transferred to a beaker.

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