



Content of Trace Elements in Serum of Patients with Carcinoma of the Larynx

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Abstract. An examination of the content of arsenic, nickel, copper, selenium, zinc, and iron in the serum of 78 patients with carcinoma of the larynx was carried out. The patients were divided into 4 groups: I – patients before treatment, II – patients after surgical treatment, III – patients after radiotherapy, IV – patients after combined treatment (surgery treatment + radiotherapy). The control group was formed of 17 patients operated for deviation of the nasal septum. Higher concentrations of arsenic, nickel and copper were found in the serum of the patients with carcinoma of the larynx before treatment (group I) as compared with the control group, whereas the concentrations of selenium, zinc and iron were lower. In the groups of patients after treatment, the highest concentrations of iron and zinc were found after surgical treatment. The level of selenium in all groups of patients was considerably lower than in the control group.

Key words: cancer; larynx; trace elements.

Introduction

Depending on their concentrations, a number of microelements show the property of either inducing or inhibiting the growth of spontaneous or chemically induced tumors¹⁹. According to the International Agency for Research on Cancer, arsenic, beryllium, cadmium, nickel and chromium have been considered human carcinogens⁴. Cobalt and copper have been considered possibly carcinogenic^{4, 5, 6, 25, 26}. Some authors also include iron²⁴. On the other hand, according to PFEIFFER and BRAVERMAN²¹, some elements, such as selenium, zinc, molybdenum and manganese, show anticarcinogenic properties.

The concentrations of these metals in serum are quite often examined in various types of carcinomas. There is, however, relatively little literature on the content of trace elements in the serum of patients with carcinoma of the larynx.

LAJTMAN et al.¹⁶ and SENGAS et al.²⁶ stated some differences in serum concentrations of such individual trace elements as selenium, zinc and copper in patients suffering from larynx carcinoma as compared with a control group. DURAK et al.⁷ observed an increased content of iron, zinc, magnesium, phosphorus in laryngeal tissues with cancer and suggest that the changed element status of cancerous larynx tissues may arise from increased requirements of cancer tissues for these elements. However the impact of trace elements on the course and initiation of neoplasms, including carcinoma of the larynx, is still unclear.

The aim of the present work is to measure and compare with a control group the concentrations of selected trace elements in the serum of untreated patients with carcinoma of the larynx and the concentrations of the same trace elements in the serum of patients before and after treatment.

Materials and Methods

Serum was taken from 78 patients with carcinoma of the larynx treated in the Clinic of Otolaryngology of the University Medical School in Wrocław. The control group consisted of 17 patients treated in the same clinic for deviation of the nasal septum. Patients of the control group and patients with carcinoma of the larynx were males and smokers. The ages of the patients were from 32 to 88 years, with average of 54.

The concentrations of the following 6 elements: arsenic (As), nickel (Ni), copper (Cu), selenium (Se), zinc (Zn) and iron (Fe) were determined according to atomic absorption spectrophotometry¹¹.

The patients were divided into the following groups: group 0 – the control group with 17 patients, group I – patients before treatment, consisting of 34 patients with a clinically diagnosed and histopathologically confirmed carcinoma of the larynx. Twenty one patients had changes of the 4th degree, 8 of the 3rd degree, 1 of the 2nd and 4 the 1st. Clinical advancement of carcinomas was assessed according to TNM classification⁸, group II: patients after an operation (laryngectomy or cordectomy) – 16 patients, 6 with the changes of the 4th degree and 3 the 3rd had had a laryngectomy, the remaining 5 the 1st degree and 2 with the 2nd, had had a cordectomy. In this group blood samples were taken for examination before the operation and 6 weeks after. Group III: patients after radiotherapy – 12 patients, 10 exhibiting first degree and 2 second degree changes. For these patients, X-ray therapy was the method of choice. In this group blood samples were taken for examination before treatment and 6 weeks after the irradiation was completed. Group IV: patients after combined treatment (laryngectomy + radiotherapy) – 16 patients, 14 with changes of the 4th degree underwent both laryngectomy and X-ray therapy, 2 patients with changes of the 2nd degree had cordectomy and X-ray therapy. From these patients blood samples were taken for examination before treatment and 6 weeks after the irradiation was completed.

Blood samples of the control group and the study groups before and after treatment were taken after fasting. After coagulation, the blood was centrifuged at 1500 rpm and the serum was separated in a water bath till it was dry. The sample was dried at 130–140°C until its weight was stable. Then concentrated nitric acid, spectrally pure POCH, was added to the sample (0.8 ml of acid for 0.1 g of the dry content) and it was heated over a reflux condenser at the boiling point of the acid (125–130°C) until nitric oxides were no longer emitted

(5–6 h). After cooling, doubly distilled water was added to obtain a total volume of 5 ml and the trace elements were determined by means of a BAIRD plasma spectrometer¹¹. For every series of measurements a control test was carried out using analogous amounts of nitric acid diluted in doubly distilled water with a total volume of 5 ml. All the results of microelement serum concentrations in the examined samples are expressed in ppm, i.e. µg/ml.

The results were statistically analyzed to obtain the arithmetic means, standard deviations, mean errors and confidence intervals. To compare the values of the calculated parameters, the F test (Fischer variance analysis) and the LSD (least significant difference) test were used.

Results

The results are presented in Tables 1 and 2. The concentrations of As, Ni and Cu were significantly higher in the serum of patients with carcinoma of the larynx before treatment (group I) than in the control group. The differences between these results are statistically significant (Table 1).

The concentration of Ni in groups I and II was significantly higher than in the control group. In groups III and IV the concentration of Ni was similar to that of the control group. In all the study groups, before and after treatment, the concentration of Cu was significantly higher than in the control group. Of all the groups after treatment, the lowest concentration of Cu was in group IV. The concentrations of Se, Zn and Fe were lower in the serum of the patients with carcinoma of the larynx before treatment (group I) than in the control group. In the case of Fe, the highest concentration was in group II, i.e. after surgery, but this result was not statistically significant. The level of Se was significantly lower in all study groups as compared with the control group. The concentration of Zn in group II (after surgery) was significantly higher than in the control group. In groups III and IV the levels of Zn were similar to the control group's.

When analyzing the concentrations of individual trace elements in the patients with carcinoma of the larynx, any correlation in the behavior of these metals was sought. It was with this purpose in mind that the correlation coefficients, presented in the Table 2, were calculated. As the table shows, there is a positive correlation (Kr) between the content of Zn and Fe (Kr – 0.5070) and Zn and Se (Kr – 0.5789), while there is a negative correlation between Se and Cu or As.

Table 1. The arithmetic means for the selected trace elements in the study groups

Groups	As (ppm)	Ni (ppm)	Cu (ppm)	Fe (ppm)	Se (ppm)	Zn (ppm)
0	0.0601	0.0358	0.3917	11.7151	0.6719	1.0248
I	0.1385	0.3374	1.7243	7.9223	0.2538	0.5119
II	0.1249	0.3837	2.1379	16.2733	0.3574	1.5317
III	0.1586	0.0014	2.1843	8.4473	0.2675	1.0552
IV	0.1166	0.0259	1.1370	9.5560	0.3251	1.0163
p(0-I)	<0.05	<0.05	<0.05	NS	<0.05	<0.05
p(0-II)	<0.05	<0.05	<0.05	NS	<0.05	<0.05
p(0-III)	<0.05	NS	<0.05	NS	<0.05	NS
p(0-IV)	<0.05	NS	<0.05	NS	<0.05	NS

NS – non-significant, $p > 0.05$.

Group 0 – the control group, 17 patients, group I – 34 patients with a clinically diagnosed and histopathologically confirmed carcinoma of the larynx, group II – 16 patients after operation (laryngectomy or corpectomy), group III – 12 patients after radiotherapy, group IV – 16 patients after combined treatment (laryngectomy + radiotherapy).

Table 2. Correlation factors between elements tested

	Zn	Fe	Ni	Cu	As
Fe	0.5070				
Ni	-0.1523	-0.0949			
Cu	0.0440	-0.1329	0.2725		
As	-0.1060	-0.0742	0.1063	0.2103	
Se	0.5789	0.0240	-0.0973	-0.4040	-0.3967

Discussion

There are relatively few reports on the content of trace elements in the serum of patients with carcinoma of the larynx. It is very difficult to compare the exact values for each elements, as the available literature shows large variations in the results obtained. It makes more sense to compare the differences between the cases investigated and control groups than to compare absolute values. The variance in the results is due both to problems of analysis (these elements are scarce in body fluids) and to the pollution of the natural environment with metals, which affects the levels of not only unnecessary, but vital trace elements as well²².

The interpretation of the results for arsenic raises no question. In our studies the patient groups showed a higher level of this element, with no differences between patients before and after treatment. Similar results were also obtained by other authors, although for other kinds of carcinoma¹⁷.

In the case of Ni, a statistically significant increase in its concentration in the serum of patients with carcinoma of the larynx before treatment and in the group of patients after surgery was detected. In other groups the level of Ni was similar to that of the control group. There are some reports concerning an increase of the risk of neoplasm occurrence among workers persistently

exposed to Ni and its compounds^{2, 28}. However, there is a lack of reports on Ni content in the serum of the patients suffering from carcinoma of the larynx and other neoplasms.

The determination of the level of Cu showed a statistically significant increase in the concentration of this element in the serum of patients in all groups studied. In the group of patients with larynx carcinomas after combined treatment, the level of Cu was lower than in the group of patients before treatment, of those after surgical treatment, and of those after radiotherapy, but slightly higher than in the control group. These results could suggest that the combined treatment is the quickest way to reach a Cu level similar to that of the control group. These observations are in agreement with research performed by other authors^{6, 25, 26}. ZAKRZEWSKI et al.²⁹ found that 47 out of 67 patients with carcinoma of the larynx exhibited normal levels of Cu in serum, 10 of them higher, and 14 lower level. Similar results were obtained by SCHWARTZ²⁴.

The results obtained in this study show a decrease in the concentration of iron in the serum of patients before treatment as compared with the control group, but this result was not statistically significant. It is striking that there were higher levels of Fe among patients after surgical treatment as compared with those of the control group. It seems that this might be due to multiple blood transfusions during and after the operation. In the blood of patients after combined treatment and after radiotherapy, the concentration of Fe was lower than in the control group and slightly higher than among patients before treatment. On the whole, these results are consistent with those reported by other authors^{12, 13, 29}. Among others, ZAKRZEWSKI et al.²⁹ examined 67 patients with carcinoma of the larynx in different degrees of clinical advancement, finding that 56,

i.e. 83%, had levels of Fe in their serum lower than normal. ZAKRZEWSKI et al.²⁹ did not notice any considerable changes in the level of iron after surgical treatment or after radiation therapy. Similar results were obtained by HOFFMAN and NIYOGI¹³ and HOFFMAN¹², who also noticed normal levels of iron in the serum of patients who had recovered after carcinoma of the larynx. A higher concentration of Fe for patients after surgical treatment and a slightly higher level of this element for other groups observed in the present paper confirm the findings of HOFFMAN and NIYOGI¹³ and HOFFMAN¹² about Fe content gradually normalizing as the patients recover.

Selenium is one of the elements to which anticarcinogenic properties are attributed²³. According to epidemiological research in the USA and Finland, individuals with low levels of Se are more likely to develop cancer. The analysis of a random sample of the population between in the ages of 25–29 demonstrated that in all the cases of deaths caused by carcinoma, the content of Se was significantly lower²⁰. On the other hand, LAJTMAN et al.¹⁶ has proposed that the lower level of Se in patients with carcinoma of the larynx is a result rather than a cause of cancer. In the present work the level of Se was significantly lower in all studied patient groups in comparison to control group. This is in agreement with the observations by other authors concerning its lower concentration in the serum of patients with other types of carcinoma¹. The data collected so far suggest that high Se level is crucial in reducing the risk of carcinoma, especially of the digestive and respiratory tracts⁹. This has resulted in the development of research on Se preparations used as chemopreventive factors in the prophylaxis of certain carcinomas⁹.

The examination of Zn content in the serum of patients before treatment (group I) showed a lower level as compared with the control group. It was considerably higher for all groups of patients after treatment (group II, III and IV). The concentration of Zn in group II (after surgery) was significantly higher than in the control group. In groups III and IV the levels of Zn were similar to the control group's. The results obtained correspond to observations by other authors. GAROFALO et al.¹⁰ examined the levels of Zn and Cu as well as the Cu-Zn ratios in 50 patients with carcinoma of the oral cavity, throat and larynx, finding that there was a tendency toward lower levels of Zn in these cases. SENGAS et al.²⁶ found a considerable drop of Zn levels in the serum of 30 patients with carcinoma of the larynx as compared with the control group. JACOBS¹⁴ and JACOBS et al.¹⁵ suggested that a greater concentration of Zn reduces the size of squamous cell carcinomas. So the

theory of the inhibiting role of Zn in carcinogenesis seems to be justified, and it has been confirmed by the results obtained in the present work.

While examining the behavior of microelements in carcinoma of the larynx, one should take into consideration mutual correlations among these elements. On the basis of the research carried out and the calculations made, one cannot draw general conclusions yet because of the small number of patients in the groups for this kind of research. However, these results clearly confirm the reports by other authors of mutual interactions between metals, which take place as early as during intestinal absorption¹⁸.

Undoubtedly there is a great need for more advanced research on the role of trace elements in carcinomas. Their importance as prognostic factors for the study of carcinomas is still a subject requiring ample documentation and further research.

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