Journal of Elementology ISSN 1644-2296

Demir D.C., Demir H., Bozan N., Belli S., Demir C. 2023.
Determination of some trace elements and heavy metal levels (Cu, Mn, Mg, Fe, Zn, Co, Pb, and Cd) in blood serum of patients with lip and oral cavity cancers.
J. Elem., 28(1): 89-106. DOI: 10.5601/jelem.2022.27.3.2315

RECEIVED: 18 July 2022
ACCEPTED: 11 January 2023

ORIGINAL PAPER

DETERMINATION OF SOME TRACE ELEMENTS AND HEAVY METAL LEVELS (Cu, Mn, Mg, Fe, Zn, Co, Pb, AND Cd) IN BLOOD SERUM OF PATIENTS WİTH LIP AND ORAL CAVITY CANCERS*

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Abstract

This study investigated copper (Cu), manganese (Mn), magnesium (Mg), iron (Fe), zinc (Zn), cobalt (Co), lead (Pb) and cadmium (Cd) levels in the blood serum of patients with lip and oral cavity cancer. Our study aims to determine the relationship of some trace elements and heavy metals with lip and oral cavity cancer. Blood serum results of 21 individuals with lip and oral cavity cancer were compared with those in the control group consisting of 30 volunteer and healthy subjects. The serum levels of trace elements and heavy metals (Cu, Mn, Mg, Fe, Zn, Co, Pb and Cd) were determined by atomic absorption spectrophotometry at Yuzuncu Yil University Central Research Laboratory (Spectrometer: Thermo Scientific C103500100, China). In this study, when the descriptive statistics and comparison results for Cu, Mn, Mg, Fe, Zn, Co, Pb, and Cd were examined, the difference between the patient and healthy group means was found to be statistically significant (p<0.05). According to these results, Zn, Fe, Mn, Mg, and Cu levels which are essential trace elements for the human body, were significantly lower in patients than in healthy groups. On the other hand, Cd, Co, and Pb levels which are harmful and toxic to the human body were significantly higher in patients than in healthy persons (p<0.05). In conclusion, deficiencies of some trace elements, such as Cu, Mn, Fe, Zn, and Co, were

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* This study was supported by Van Yuzuncu Yil University BAPB (Scientific Research Projects Department) as Project No: FYL-2017-6088.

detected in the blood serums of patients with lip and oral cavity cancer. Deficiencies of trace elements in patients with lip and oral cavity cancer may result from excessive nutrient consumption or malnutrition of cancer patients, in addition to which trace element and heavy metal levels (Cu, Mn, Mg, Fe, Zn, Co, Pb and Cd) can affect the etiopathogenesis of the disease.

Keywords: lip, oral cavity cancer, trace element, heavy metal

INTRODUCTION

Cancer, which is defined as uncontrolled division of cells due to the disruption of genes controlling cell death, proliferation and differentiation, is a multistep, multifactorial pathological disease. It has always been important in the scientific and medical communities due to such reasons as very high incidence and mortality, low five-year survival rates after the disease, high rate of disability, and lengthy and expensive treatment (Lichtenstein et al. 2000). Cancer cells, whose reproduction is unlimited and uncontrolled, make room for themselves and enlarge the area they occupy by phagocytizing healthy cells through secreting toxic and proteolytic enzymes. As a result, they not only prevent the proper functioning of tissues and organs in which the disease started, but also affect adversely the functions of distant tissues and organs through metastasis (Lichtenstein et al. 2000).

Head and neck cancers (HNC) constitute 6% of all cancer types and are in the sixth place in incidence. Worldwide, approximately 900,000 people are diagnosed with HNC annually, and more than 400,000 die due to this cancer (IARC, 2021). Cancers in the head and neck region can be listed as tumors of the skin, nasal cavity, paranasal sinuses, nasopharynx, lip, oral cavity, oropharynx, larynx, hypopharynx, and cervical esophagus, cervical trachea, neck, salivary glands, parapharyngeal region. Biological behavior, clinical course, and response to the treatment applied in these cancers differ (López et al. 2016).

Oral cavity cancer is a significant cause of morbidity and mortality worldwide. The most common histological type of cancer in the oral cavity is oral squamous cell carcinoma (OSCC), which represents over 90% of all oral cancers. Tobacco and alcohol are the main etiological risk factors of OSCC. The most common cancers in oral cavity cancers are those located in the lower region, such as the oral tongue (51%), followed by cancers of the floor of the mouth (15%) and lower gum cancers (14%) – Zanoni et al. (2019).

Trace elements, which are sufficient for the body in very small amounts, constitute less than 0.02% of the total body weight. The daily amount of trace elements required for the body is between 15-80 μg. Deficient supply of trace elements under these amounts creates various metabolic problems while their excess may give rise to toxic effects (Emre et al. 2013). Despite these small quantities in which they should occur, trace elements have varied and critical biological functions. Among these tasks, they have antioxidant effects, act as cofactors for some enzymes, are a stabilizer for cell membranes, assist in hormone activities, participate in anabolism, incorporate metalloenzymes and metalloproteins into structural components, protect against minerals that are toxic to metabolism, help transport some substances in the circulatory system, contribute to the repair of wounds and tissue damage, and accelerate learning. In addition to this, they are involved in the transmission of small electrical stimuli to the cardiac tissues, which enables the functioning of the heart muscle, the realization of brain functions, and the adjustment of the water pressure of the cells (Çavuşoğlu et al. 2008, Emre et al. 2013).

People are constantly exposed to a large number of toxic elements that are dispersed in nature. Lead, cadmium, cobalt and other toxic trace elements, show poisonous effects on people even when present at low levels (Morais et al. 2012). Heavy metals are classified as essential and non-essential ones. Essential heavy metals in their living forms act as cofactors in enzymatic reactions. However, they show toxic effects after surpassing a certain concentration (1-10 ppm: part per million) (Fe, Cu, Zn, Ni and Se). In contrast, non-essential heavy metals (Hg, Cd and Pb) are toxic even at low concentrations (0.001-0.1 ppm) – Rzymski et al. (2015).

Cu is a trace element necessary for developing and replicating all eukaryotic cells. It must be obtained from external sources as metabolic activities cannot create or destroy it (Nevitt et al. 2012). It is essential in various biochemical reactions as a cofactor for superoxide dismutase (SOD), which plays a vital role in protecting the body against free radicals (Zhang et al. 2018). It is also an essential trace element that acts as a critical component in several copper-dependent enzymes that regulate processes such as cell proliferation and angiogenesis (Lopez et al. 2019). However, if Cu is found in high concentrations in metabolism, it creates a potentially toxic effect (Shanbhag et al. 2021).

Mn is an essential nutrient that is necessary for the functioning of the superoxide dismutase enzyme system and therefore functions as a coenzyme in various biological processes. Mn SOD, which also contains Mn and is in the tetrameric form, is found in mitochondria. The processes it is involved in include macronutrient metabolism, bone formation, free radical defense systems, ammonia scavenging in the brain, and neurotransmitter synthesis. It is a critical component of many proteins and enzymes in all tissues. However, excessive exposure to environmental Mn through potable water contaminated with industrial waste can lead to toxic effects and neurological disorders because of the accumulation of Mn in the brain (Erikson, Aschner 2019).

Mg is the second most abundant cation in the cell, involved in almost all metabolic and biochemical processes. Mg, which acts as a cofactor in many enzymatic reactions, plays a role in numerous biochemical events such as protein and nucleic acid synthesis, regulation of metabolic pathways, neuronal transmission, neuromuscular function and regulation of heart rhythm, and preservation of electrolyte balance (Barbagallo et al. 2021).

Fe is an element of vital importance in human metabolism. It acts as both an electron donor and electron acceptor. It is required for oxygen transport and storage by hemoglobin, as well as for muscle oxygenation by myoglobin, cellular respiration by proteins of the mitochondrial electron transport chain, and DNA synthesis by ribonucleotide reductase. Fe also participates in various enzymatic reactions in the cytosol and other cellular divisions. Owing to its ability to receive and transfer electrons, Fe may cause oxidative stress and tissue damage. Since iron is essential in energy metabolism and vital in preventing potential harm to cells and tissues, its absorption, transfer and metabolism are strictly regulated (Galaris et al. 2019).

Zinc, a trace element essential for cellular metabolism, is required for the immune function, wound healing, blood clotting, thyroid function, as well as the functioning of numerous transcription factors and over 300 enzymes. Its deficiency causes an increase in oxidative stress, various metabolic diseases, endocrine disorders, developmental problems, neurodegenerative problems, immunodeficiencies, inflammatory pathologies, skin diseases, and delayed wound healing (Lubiński et al. 2021).

Copper, widely available in nature, appears in many anthropogenic activities. Copper also has an important biological role, as it is located as the central atom in the middle of the porphyrin ring of vitamin B12, and is necessary for the biological activity of this vitamin. However, it has been observed in many different studies that excessive Co exposure causes various health problems, including neurological (e.g. hearing and visual impairment), cardiovascular and endocrine deficiencies (Bernianti et al. 2020).

Lead is an essential element for the body; contrarily, it is quite dangerous for human health. Pb is a toxic element that is usually released from industrial waste and combustion of fossil fuels. The primary sources of anthropogenic transmission of lead are industries, gasoline production and use, as well as paints. The most common form of transmission in developed countries is occupational exposure. By inhibiting the enzymes involved in hemoglobin synthesis, Pb prevents adequate oxygen and blood transport to the tissues and thus creates a toxic effect in the body (Rashed 2006). If the lead level in the blood reaches 400 μg L^{-1}, anemia begins. In lead poisoning, the share of the erythrocyte-protoporphyrin complex in the blood increases because lead inhibits the formation of complexes of the iron element with protoporphyrin-IX in the blood. Health problems caused by Pb exposure, which is very harmful, especially for children, include brain and nervous system damage, gastrointestinal problems, anemia, liver and kidney damage, infertility, and developmental delays (Rhee et al. 2021).

Cd, which is very harmful to humans, accumulates in the body over the years and remains in it for a very long time, even at shallow exposure (Naja, Volesky 2009). It can be taken into the body through inhalation, nutrition, cigarette smoke, and drinking water contaminated with Cd. Approximately 5% of Cd is taken into the human body with nutrients and is mainly absorbed in the lungs. The primary source of Cd absorbed in the lungs is smoking. Inhalation of excessive amounts of Cd can cause lung damage and death. In addition, vomiting and diarrhea result from excessive intake of Cd with nutrients. Cd taken in low amounts for a long time through the air, water and nutrients accumulates in the kidneys and may cause death because it will disrupt the renal arterioles and glomeruli (Naja, Volesky 2009, Satarug et al. 2010). It has been determined that mortality rates increase in humans due to cadmium pollution, detected in the urine excreted with glomerular protein and in the structure of kidney stones (Satarug et al. 2010).

Trace element levels in the human body change in many diseases and the course of an illness will change depending on the above changes. In this study, changes in Cu, Mn, Mg, Fe, Zn, Co, Pb and Cd levels were investigated in patients with lip and oral cavity cancer and healthy individuals. The aim of the study is to determine trace element and heavy metal levels in lip and oral cavity cancers and their relationship with the disease. It is thought that the results obtained will shed light on other findings regarding lip and oral cavity cancer.

MATERIAL AND METHOD

In this study, a total of 21 lip and oral cavity cancer patients aged 18-65 years were diagnosed and followed in the Department of Otorhinolaryngology, Faculty of Medicine, Yüzüncü Yıl University and Otorhinolaryngology Clinic of Bağcılar Training and Research Hospital of the University of Health Sciences. Their blood samples were taken between June 2017 and January 2018. Fourteen of the patients were male, and seven were female. Of the male patients, six were smokers, and four were using alcohol. Of the female patients, two were smokers, and one was using alcohol. All cancer patients underwent histopathological examinations and cancer disease was confirmed. There was no age restriction in the patients included in the study. In addition, the patients did not have any other chronic or metabolic diseases. The inclusion criteria for the group of healthy persons were: non-smokers, abstaining from alcohol, with no chronic or metabolic diseases, not taking vitamin supplements, and to being in the same age range as the patients studied. Before the blood samples were collected, the Approval of the Local Ethics Committee of Clinical and Laboratory Researches of Van Training and Research Regional Hospital had been obtained. To compare the blood test results obtained from individuals with lip and oral cavity cancer, 30 healthy volunteers whose ages were in the same range as those with lip and oral cavity cancer patients were used as a control group.

In this study, a sample of 4 ml of blood was taken each individual with lip and oral cavity cancer and from each healthy person in the control group, and placed in a biochemistry tube. The blood samples were centrifuged for 5 min at 4000 rpm in a Nüve NF 800 brand centrifuge. Then, the serum was separated and kept at -20°C until the day of processing. When the target number of samples was reached, all samples were brought to room temp. (15-18°C) by carefully mixing and swirling before starting the study. Then, 1 ml of serum was taken into falcon tubes. 1 ml of 20% TCA was added to it. The tubes were closed and vortexed. They were kept in an oven at 90°C for 15 minutes. They were then allowed to cool, and when cooled, centrifuged. The resulting supernatant was taken into clean tubes (Olson, Hamlin 1969). The serum levels of trace elements and heavy metals (Zn, Cu, Mg, Pb, Mn, Cd, Co, and Fe) were determined by the atomic absorption spectrophotometery (AAS) method in the NEU Center Research Laboratory (Spectrometer: Thermo Scientific C103500100, China). AAS is an instrumented analysis method that adopts the principle of absorbing the sent electromagnetic rays of the element atoms gasified by increasing the temperature. The method is based on the principle of calculating the absorbed beam according to the Lambert-Beer law. According to this method, the atom or substance passes from the basic energy level to the excited energy level by absorbing the appropriate wavelength in the transmitted beam. The atom, which is in an excited state for 10^{-6} and 10^{-9} seconds, then returns to its original state by giving back the absorbed beam (Risby 2006). Wavelengths of the elements are: Cu:324.8, Mn:403.1, Mg:285.2, Fe:248.3, Zn:213.9, Co:312.8, Pb:325.8, Cd:254.5

Statistical analysis

Descriptive statistics for the features under consideration were expressed as mean and standard deviation. The *t*-Test was used in the cases where normal distribution condition was provided in paired group comparisons, and the Mann-Whitney *U* test statistic was used in cases where normal distribution condition was not satisfied. The SPSS statistical package program was used for the calculations, and the statistical significance level was calculated as 0.05.

RESULTS

Blood samples from 21 individuals with lip and oral cavity cancer were compared with the blood samples in the control group of 30 voluntary and healthy individuals.

Descriptive statistics and comparison results for Cu, Mn, Mg, Fe, Zn, Co, Pb and Cd concentrations in serum are given in Table 1. According to these

Table 1

Trace elements in patients with lip and oral cavity cancer and healthy persons

Parameter	Healthy group n=30 Mean±SEM (min-max)	Patient group n=21 Mean±SEM (min-max)	P value
Cu mg L^{-1}	2.35±0.33 (0.03-2.92)	0.71±0.18 (0.41-1.017)	0.001
Mn mg L^{-1}	0.13±0.01 (0.11-0.14)	0.03±0.01 (0.02-0.05)	0.001
Mg mg L^{-1}	39.81±1.62 (37.1-40.09)	25.30±1.24 (21.83-27.06)	0.001
Fe mg L^{-1}	4.90±0.18 (.4.57-5.10)	1.25±0.58 (0.33-2.31)	0.001
Zn mg L^{-1}	2.56±0.24 (2.25-22.97)	0.78±0.29 (0.32-1.42)	0.001
Co mg L^{-1}	0.02±0.004 (0.01-0.02)	0.08±0.02 (0.04-0.12)	0.001
Pb mg L^{-1}	0.06±0.01 (0.05-0.09)	0.22±0.10 (0.13-0.40)	0.001
Cd mg L^{-1}	0.049±0.02 (0.02-0.08)	0.31±0.16 (0.10-0.82)	0.001

data, the difference between the means of the patient and control groups was statistically significant for all trace elements and heavy metals ($p<0.05$).

While the Cu level was 2.35±0.33 mg L^{-1} in the healthy control group, it was found as 0.71±0.18 mg L^{-1} in the patients with lip and oral cavity cancer. The difference between the two groups was statistically significant ($p<0.05$).

Mn level was statistically significantly higher in the healthy control group (0.13±0.01 mg L^{-1}) compared to the patients with lip and oral cavity cancer (0.03±0.01 mg L^{-1}) – $p<0.05$.

The concentration level of Mg element was 39.81±1.62 mg L^{-1} in the healthy control group and 25.30±1.24 mg L^{-1} in patients with lip and oral cavity cancer. The difference between the two groups was statistically significant ($p<0.05$).

Fe level was statistically significantly higher in the healthy control group (4.90±0.18 mg L^{-1}) than in the patients with lip and oral cavity cancer (1.25±0.585 mg L^{-1}) – $p<0.05$.

While the Zn level was 2.56±0.24 mg L^{-1} in the healthy control group, it was 0.78±0.29 mg L^{-1} in patients with lip and oral cavity cancer. The difference between the two groups was statistically significant ($p<0.05$).

The results obtained for the heavy metals were different from those for the trace elements. According to the data in Table 1, while the Co level was 0.02±0.0043 mg L^{-1} in the healthy control group, it was found as 0.078±0.02 mg L^{-1} in the patients with lip and oral cavity cancer, and this difference between the two groups was statistically significant ($p<0.05$).

The Pb level was statistically significantly lower in the healthy control group (0.0641±0.0139 mg L^{-1}) than in the patients with lip and oral cavity cancer (0.22±0.10 mg L^{-1}) – $p<0.05$.

Cd level was statistically significantly higher in patients with lip and oral cavity cancer (0.31±0.16 mg L^{-1}) when compared to the control group (0.05±0.02 mg L^{-1}) – $p<0.05$.

DISCUSSION

In this study, we aimed to detect trace elements and heavy metal levels in patients with lip and oral cavity cancer, and to determine their relationship with the disease. In this context, we compared the findings related to trace elements and heavy metals that we studied with other studies in the literature. Since there are many studies on the subject in the literature, we could make comparisons with only a few of them. However, although there are studies on the relationship between cancer-trace elements in general in the literature, there are limited studies comparing lip and oral cavity cancer with trace elements and heavy metals.

The serum Cu levels of the patients were reported significantly higher than the healthy control groups in cases of bladder cancer (Yıldız 2015), colon cancer (Emre et al. 2013), breast cancer (Demir, Demir 2016), cervical cancer (Zhang et al. 2018), prostate cancer (Saleh et al. 2020) and lung cancer (Zhang, Yang 2018). In another study on lung cancer (Çobanoğlu et al. 2010), serum Cu levels of patients were significantly lower than in healthy control groups. In a study on ovarian cancer, it was shown that the preoperative blood levels of Cu in the patients were lower than in the control group and higher than in the postoperative group, although the differences were not statistically significant (Bilici 2014). In other studies by Baharvand et al. on oral cancer patients (Baharvand et al. 2014), by Tiwari et al. on potentially malignant disorders and oral cancer (Tiwari et al. 2016), and by Neethi et al. on oral submucosal fibrosis (Neethi et al. 2013), the serum levels of Cu of the patients were significantly higher. In cases of oral submucous fibrosis and oral squamous cell carcinoma (Khanna, Karjodkar 2006, Khanna et al. 2013), laryngeal cancer (Taysi et al. 2003), the serum levels of Cu of the patients were significantly higher than in the healthy control group. In our study, we found that the Cu level in the blood of patients with lip and oral cavity cancer was significantly lower than in the healthy control group (Table 1). The concentration of Cu element was 2.35±0.33 mg L^{-1} in the healthy control group and 0.71±0.18 mg L^{-1} in lip and oral cavity cancer patients. The low level of Cu, which has a role in many critical biochemical events in metabolism, in the patient group may affect the prognosis of the disease. In conclusion, the low Cu level may play a decisive role in developing the disease in lip and oral cavity cancers.

Bladder cancer (Yıldız 2015), lung cancer (Zabłocka-Słowińska et al. 2018), gastrointestinal cancer (Türkdoğan et al. 2022), prostate cancer (Saleh et al. 2020), ovarian cancer (Bilici 2014) and acute leukemia (Demir et al. 2011) were investigated in various studies, where the serum Mn levels of the patients were significantly lower than in the healthy control groups. In a study conducted on patients with chronic lymphocyte leukemia, although the serum Mn levels of the patients were lower than in the healthy control groups, the difference was not statistically significant (Gündoğdu et al. 2007). In colon cancer (Emre et al. 2013), lung cancer (Çobanoğlu et al. 2010), and prostate cancer, analyzed in different studies, the serum Mn levels of the patients were significantly higher than in the healthy control groups. In this study on the lip and oral cavity cancers, the difference between the mean levels of the patient and control groups in terms of the serum Mn levels was statistically significant, while the Mn levels in the serum of patients with lip and oral cavity cancer (0.03±0.01 mg L^{-1}) were significantly lower than in the healthy control group (0.13±0.01 mg L^{-1}) – $p<0.05$ (Table 1). The low level of Mn may affect the development of the disease in patients with cancers of the lip and oral cavity, as it plays a role in the functioning of the superoxide dismutase enzyme system, which protects cells from the carcinogenic effects of chemicals and radiation.

In a study on lung cancer (Çobanoğlu et al. 2010), the serum Mg level was found to be significantly lower than in the healthy control group, while in another study on lung cancer (Song et al. 2018) there was no statistically significant difference between the serum Mg levels in the patient and the healthy groups. A study on gallstones and gallbladder cancer reported that a high Mg level increased the risk of gallstones and reduced the risk of gallbladder cancer (Lee et al. 2020). Reis et al. proved that the presence of high bloodstream magnesium, neoplasia and sepsis tended to increase the odds ratio for mortality in critically ill patient (Reis et al. 2017). In a study conducted on patients with colon cancer, it was determined that the serum Mg levels of the patients were higher than those of healthy people (Emre et al. 2013). Bilici reported that the Mg level of patients with ovarian cancer was higher in the preoperative group compared to the control group and the postoperative group (Bilici 2014). In a study on children with acute leukemia, significantly lower Mg content was found in diseased children's serum and whole blood compared to healthy subjects (Afridi et al. 2018). Polter et al. reported that there was an inverse relationship between serum Mg levels and the risk of colorectal cancer (Polter et al. 2018). When the Mg concentration was compared with that in healthy subjects, the serum Mg levels of patients were significantly lower in both the blood plasma and saliva of oral squamous cell carcinoma patients (Aziz et al. 2018). Our study on the lip and oral cavity cancers found that the level of Mg in the blood of patients with lip and oral cavity cancer was significantly lower than in the healthy control group (Table 1). The concentration level of Mg element was 39.81±1.62 mg L^{-1} in the healthy control group and 25.30±1.24 mg L^{-1} in patients with lip and oral cavity cancer. The low level of Mg in the patient group, which has a role in many critical biochemical events in metabolism, may affect the prognosis of the disease. Since Mg plays a role in many biochemical events, its low level may be a determinant in the etiopathogenesis of lip and oral cavity cancers.

In two studies on breast cancer (Demir, Demir 2016, Rajizadeh et al. 2017), serum Fe levels in patients were significantly higher than in healthy control groups. Hou et al. recently declared that serum Fe levels were not in a relationship with the risk of breast cancer or its subtypes (Hou et al. 2021). A study of patients with early-stage triple-negative breast cancer (Hua et al. 2021) reported that patients in the high Fe group had a significantly shorter median survival than patients in the low Fe group. In a study on lung cancer patients (Chen et al. 2017), serum Fe levels were not found to be statistically significant in both women and men in different pathological types, while in another study (Sukiennicki et al. 2019), significantly higher serum Fe levels were detected in patients with lung cancer compared to healthy controls. Dayani et al. promised that excess Fe load might increase the risk of cancer formation since it directly increases the formation of hydroxyl peroxide and free hydroxyl radicals (via Fenton and Haber-Weiss reactions) – Dayani et al. (2004). In two studies on prostate cancer (Kaba et al. 2014, Saleh et al. 2020), serum Fe levels in patients were significantly higher than in healthy control groups. In the study of Bilici on ovarian cancer, it was shown that the Fe levels in patients were lower than in the control and postoperative groups (Bilici 2014). In a recent study on cervical cancer (Chen et al. 2020), patient serum Fe levels were significantly lower than healthy controls. In a study on liver cancer (Tian et al. 2021) no significant difference was found between serum Fe levels of the patient and control groups. In our study on lip and oral cavity cancers, the difference between the patient and control group mean levels in terms of Fe levels were statistically significant. Serum Fe levels of patients with lip and oral cavity cancers (1.25±0.58 mg L^{-1}) were significantly lower than the healthy control group (4.90±0.18 mg L^{-1}) – p<0.05 (Table.1). The low level of Fe in the patient group, which has a vital role in general cellular functioning and is primarily involved in energy metabolism in the organism, may affect the prognosis of the disease. This may cause problems in the energy metabolism of the cells and may lead to disruptions in many biological activities such as oxygen, electrons, enzymes, and DNA synthesis. Furthermore, in patients with lip and oral cavity cancers, low Fe levels may affect the prognosis and etiopathogenesis of the disease.

Ovarian cancer (Cunzhi et al. 2003), cervical cancer (Yaman et al. 2007, Okunade al. 2018, Xie et al. 2018), bladder cancer (Yıldız 2015), lung cancer (Çobanoğlu et al. 2010, Wang et al. 2019), prostate cancer (Kaba et al. 2014, Saleh et al. 2020) in different studies, it was found that the level of Zn in the blood serum of the patients was significantly lower than the healthy control group. A study on colon cancer determined that the zinc level was higher than in healthy people (Emre et al. 2013). Recently Lubinski et al. eveluated laryngeal cancer patients and submitted that the risk of death consistently decreased as circulating zinc levels increased (Lubinski et al. 2021). No significant difference was found between the breast cancer patient serum Zn levels and the healthy control group (El-Attar et al. 2020). In a study on patients with salivary gland tumors (benign and malignant), the difference between serum Zn levels in patients and control groups was not statistically significant (Jaafari- Ashkavandi et al. 2019). A meta-analysis (250 articles) study on oral squamous cell carcinoma reported that the serum Zn level in patients was significantly higher than in healthy people (Keshani et al. 2021). A study of 59 patients with cancer of the larynx, salivary gland, oral cavity, and tongue determined that zinc levels decreased in the hair and nails (Woźniak et al. 2012). In our study on lip and oral cavity cancers, the difference between patient and control group mean Zn levels was statistically significant. In our study, it was found that the serum Zn level of patients with lip and oral cavity cancer was significantly lower (0.78 ± 0.29 mg L^{-1}) than in the healthy control group (2.56 ± 0.24 mg L^{-1}) – $p<0.05$ (Table 1). The low level of Zn, a solid antioxidant element, in the patient group may affect the prognosis of the disease. This indicates that adequate levels of antioxidant substances do not enter the cells and that these patients are exposed to high levels of oxidative stress.

Lung cancer (Çobanoğlu et al. 2010), prostate cancer (Ozmen et al. 2006), bladder cancer (Yıldız 2015) and ovarian cancer (Bilici 2014) in different studies, it was shown that the level of Co in the blood serum of the patients was significantly higher than in the healthy control group. In another study, it was declared that osteoblast-like cells transformed into a tumorigenic phenotype in individuals exposed to cobalt and activated the expression of genes associated with cancer (Demir, Demir 2016). In a case analysis conducted on kidney cancer, it was informed that high Co levels are related to the prognosis of the disease (Massardier et al. 2020). In a study on gastrointestinal cancers, the mean serum Co level of esophageal cancer patients was significantly lower than in controls and gastric cancer patients (Türkdoğan et al. 2022). On patients at risk of gastric cancer, serum cobalt levels were higher in H. pylori-infected individuals than in non-H. Pylori--infected individuals (Hu et al. 2018). Arooj et al. reported that the mean serum cobalt levels in menstruating breast cancer patients were significantly lower than in healthy controls, and mean serum cobalt levels in premenopausal and postmenopausal patients women were significantly lower than in healthy controls (Arooj et al. 2021). A study on acute leukemia patients found no statistical difference between patient serum Co levels and healthy controls (Demir et al. 2011). In our study, it was found that the Co level in the blood of patients with lip and oral cavity cancer was significantly higher than the healthy control group (p<0.05) – Table 1. The concentration of Co element was 0.02±0.004 mg L^{-1} in the healthy control group and 0.08±0.02 mg L^{-1} in patients with lip and oral cavity cancer. The high level of Co, a heavy metal, in the patient group may affect the prognosis of the disease. In addition, high Co levels in lip and oral cavity cancers may decrease the antioxidant defense system in individuals with this disease while increasing the level of oxidative stress, which may accelerate cancer development.

Lung cancer (Çobanoğlu et al. 2010), kidney cancer (Geçit et al. 2011), renal cell carcinoma (Pirinççi et al. 2013), colon cancer (Emre et al. 2013), ovarian cancer (Bilici 2014), malignant glioma (Arslan et al. 2011), thyroid adenoma (Li et al. 2017), laryngeal papilloma and laryngeal cancer (Olszewski et al. 2006), and acute leukemia (Demir et al. 2011) In different studies, it has been reported that the Pb level in the blood serum of the patients is higher than the healthy control group. In a study on gastrointestinal cancer patients, no statistically significant difference was found between mean serum Pb levels in all (esophageal, stomach, and colon) cancer groups and controls (Türkdoğan et al. 2022). In our study on the lip and oral cavity cancers, the difference between the mean Pb levels of the patient and the control group was statistically significant, and the serum Pb level of the patients with lip and oral cavity cancer (0.22±0.10 mg L^{-1}) was significantly higher than the healthy control group (0.06±0.01 mg L^{-1}). – p<0.05 (Table 1). The high level of Pb in the patient group, which has a dangerous toxic effect on human health, may affect the progress of the disease. Therefore, high Pb levels in lip and oral cavity cancers may stimulate cancer development by boosting the level of oxidative stress in the cells.

Ovarian cancer (Bilici 2014), malignant glioma (Arslan et al. 2011), lung cancer (Çobanoğlu et al. 2010), bladder cancer (Geçit et al. 2011, Yıldız 2015), colon cancer (Emre et al. 2013), pancreatic cancer (Kriegel et al. 2006), leukemia (Khan et al. 2017) and renal cell carcinoma (Pirinççi et al. 2013) in different studies, patient serum Cd levels were found to be significantly higher than in healthy controls. In our study, the level of Cd in the blood of patients with lip and oral cavity cancer was significantly higher than in the healthy control group (Table 1). The concentration level of Cd was 0.05±0.02 mg L^{-1} in the healthy control group and 0.31±0.16 mg L^{-1} in patients with lip and oral cavity cancer. The statistical significance between the two groups was p<0.05 (Table 1). A significant heavy metal Cd level in the patient group may affect the prognosis of the disease. This may indicate an excess of Cd entering the cells. Furthermore, a high Cd level in the lip and oral cavity can accelerate cancer development by enhancing the level of oxidative stress in cells.

CONCLUSION

In conclusion, Zn, Fe, Mn, Mg, and Cu levels, essential trace elements for the human body, were significantly lower in patients than in healthy control groups. On the other hand, Cd, Co, and Pb levels, which have harmful and toxic effects on the human body, were significantly higher in patients than in healthy control groups. According to these results, trace elements and heavy metal levels in the lip and oral cavity can affect the prognosis and etiopathogenesis of the disease. However, in various studies on cancer, serum Cu, Mn, Mg, and Fe levels were lower compared to healthy individuals and higher in some studies. No statistically significant difference was found in some studies. Therefore, more comprehensive studies on the subject are needed. In most studies on Zn, a critical antioxidant element, the Zn level in the blood serum of cancer patients was low. Therefore, it was concluded that the serum Zn level could be accepted as an essential biomarker for cancer. In addition, it was considered that zinc may be an important and determining factor for the pathogenesis of lip and oral cavity cancer and may affect the prognosis of the disease. We think that the serum Pb and Cd levels are essential biomarkers for cancer, since the levels of Pb and Cd determined in patient sera in this study are in parallel with different studies on cancer in the literature. On the other hand, Co was high in some of the studies on cancer, low in some, and did not show a significant difference in some, hence further studies are needed.

Deficiencies of trace elements in patients with lip and oral cavity cancer may result from excessive consumption of nutrients or malnutrition of cancer patients. Nutritional therapy rich in trace elements should be considered as one of the cancer-fighting strategies. The disadvantage of the study is the small number of patients admitted to a hospital serving patients in a local area. For this reason, we think that it would be beneficial to conduct a study covering a larger area and with a more significant number of patients. We think that this study will benefit future studies on lip and oral cavity cancers.

Data availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

ACKNOWLEDGEMENT

We would like to express appreciation to Van Yüzüncü University and Medical Faculty Research Hospital for their assistance given for this research.

This study was supported by VAN YYÜ BAPB (Scientific Research

Projects Department) as Project No: FYL-2017-6088. We would like to thank VAN YYÜ BAPB for their contributions.

Ethics declarations

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The study was approved by the Local Ethics Committee of Clinical and Laboratory Researches of Van Training and Research Regional Hospital (Registration number: B.30.2.YYU.0.01.00.00/112; Date of registration; 15/11/2017).

Conflict of interest

The authors reported no potential conflict of interest.

This study is adapted from the thesis work "Determination of some trace elements and heavy metal level (Cu, Mn, Mg, Fe, Zn, Co, Pb, and Cd) in blood serum of patients with lip and oral cavity cancers".

REFERENCES

Afridi H.I., Kazi T.G., Talpur F.N. 2018. *Correlation of calcium and magnesium levels in the biological samples of different types of acute leukemia children.* Biol. Trace Element Res., 186: 395-406. DOI: 10.1007/s12011-018-1340-z

Arooj B., Ahmed S., Saleem M., Khurshid R., Zia M. 2012. *Serum trace elements in diagnosis of breast malignancy.* J. Ayub Med. Coll. Abbottabad., 24(2): 62-4. PMID: 24397055

Arslan M., Demir H., Arslan H., Gokalp A.S., Demir C., 2011. *Trace elements, heavy metals and other biochemical parameters in malignant glioma patient.* Asian Pac. J. Cancer Prev., 12(2): 447-451.

Aziz N.Z., Arathi K., Prasad B. G., Desai D., Shetty S.J., Shahid M. 2018. *Evaluation of magnesium levels in blood and saliva of oral squamous cell carcinoma and potentially malignant disorders by xylidyl blue method.* J Oral Maxillofac Pathol, JOMFP, 22(1): 147. https://doi.org/10.4103/jomfp.JOMFP_34_17

Baharvand M., Manifar S., Akkafan R., Mortazavi H., Sabour S. 2014. *Serum levels of ferritin, copper, and zinc in patients with oral cancer.* Biomed. J., 37(5): 331-6. DOI: 10.4103/2319-4170.132888 PMID: 25179706

Barbagallo M., Veronese N., Dominguez L.J. 2021. *Magnesium in aging. Health and diseases.* Nutrients, 13(2): 463. https://doi.org/10.3390/nu13020463

Berniyanti T., Palupi R., Kriswandini I.L., Bramantoro T., Putri I.L. 2020. *Suitability of MDA, 8-OHdG and wild-type p53 as genotoxic biomarkers in metal (Co, Ni and Cr) exposed dental technicians: a cross-sectional study.* BMC Oral Health, 20(1): 65. https://doi.org/10.1186/s12903-020-1049-1

Bilici M. 2014. *Investigation of trace element levels (Zn, Cu, Mg, Pb, Mn, Cd, Co and Fe) in blood before and after surgery in patients with ovarian cancer.* PhD Thesis, T.C Yüzüncü Yıl University, İnstitute of Natural and Applied Sciences, Department of Chemistry, Van, Turkey.

Chen S., Shen L., Luo S., Lan X., Wang, L. 2020. *Association between serum iron levels and the risk of cervical cancer in Chinese: a meta-analysis.* J Int Med Res, 48(3): 300060519882804. https://doi.org/10.1177/0300060519882804

Chen Z., Zhu B., Ou C., Li Y. 2017. *Serum ferritin and primary lung cancer.* Oncotarget, 8(54): 92643-92651. https://doi.org/10.18632/oncotarget.21518

Cunzhi H., Jiexian J., Xianwen, Z, Jingang G., Shumin Z., Lili D. 2003. *Serum and tissue levels of six trace elements and copper/ zinc ratio in patients with cervical cancer and uterine myoma.* Biol. Trace Elem. Res., 94: 113-122.

Çavuşoğlu K., Çakır Arıca Ş., Kurtman Ç. 2008. *Determination of changes in plasma trace element levels of lung cancer patients receiving radiotherapy* F.Ü. Sağ. Bil. Derg. 22(4): 211-222. (in Turkish)

Çobanoğlu U., Demir H., Sayır F., Duran M., Mergan D. 2010. *Some mineral, trace element and heavy metal concentrations in lung cancer.* Asian Pac. J. Cancer Prev., 11: 1383-1388

Dayani P.N., Bishop M.C., Black K., Zeltzer P.M. 2004. *Desferoxamine (DFO)-mediated iron chelation: rationale for a novel approach to therapy for brain cancer.* J. Neurooncol., 67: 367-377.

Demir C., Demir H., Esen R., Sehitogullari A., Atmaca M., Alay M. 2011. *Altered serum levels of elements in acute leukemia cases in Turkey.* Asian Pac. J. Cancer Prev., 12(12): 3471-3474. PMID: 22471499

Demir H., Demir C. 2016. *Effect of heavy metal and some trace element levels on radiotherapy taken breast cancer patients.* Med Sci Discov, 3(3): 116-119.

El-Attar E., Kamel A., Karmouty A., Wehida N., Nassra R., El Nemr M., Kandil N.S. 2020. *Assessment of serum CoQ10 levels and other antioxidant markers in breast cancer.* Asian Pac. J. Cancer Prev., 1;21(2): 465-471. DOI: 10.31557/APJCP.2020.21.2.465 PMID: 32102525; PMCID: PMC7332135

Emre Ö., Demir H., Dogan E., Esen R., Gur T., Demir C., Gonullu E, Turan N., Ozbay M.F. 2013. *Plasma concentrations of some trace element and heavy metals in patients with metastatic colon cancer.* J Cancer Ther, 4: 1085-1090.

Erikson K.M., Aschner M. 2019. *Manganese: Its role in disease and health.* Met. Ions Life Sci., 14: 19:/books/9783110527872/9783110527872-016/9783110527872-016.xml. DOI: 10.1515/9783110527872-016. PMID: 30855111

Galaris D., Barbouti A., Pantopoulos K. 2019. *Iron homeostasis and oxidative stress: An intimate relationship.* Biochim. Biophys. Acta Mol. Cell Res., 1866(12): 118535. DOI: 10.1016/j.bbamcr.2019.118535

Gecit I., Kavak S., Demir H., Güneş M., Pirinçci N., Cetin C., Ceylan K., Benli E., Yildiz I. 2011. *Serum trace element levels in patients with bladder cancer.* Asian Pacific J Cancer Prev, 12(12): 3409-3413.

Gundogdu M., Kaya H., Gulcin I., Erdem F., Cayir K., Keles M., Yilmaz A. 2007. *Oxidase activity of ceruloplasmin and some acute phase reactant and trace element concentrations in serum of patients with chronic lymphocytic leukemia.* Scott. Med. J. 52(1): 24-7. DOI: 10.1258//rsmsmj.52.1.24. PMID: 17373421

Hou C., Hou Q., Xie X., Wang H., Chen Y., Lu T., Wu Q., Liang Y., Hu Y., Mao Y. 2021. *Serum iron status and the risk of breast cancer in the European population: a two-sample Mendelian randomisation study.* Genes Nutrition, 16(1): 9. https://doi.org/10.1186/s12263-021--00691-7

Hu A., Li L., Hu C., Zhang D., Wang C., Jiang Y., Zhang M., Liang C., Chen W., Bo Q., Zhao Q. 2018. *Serum concentrations of 15 elements among helicobacter pylori-ınfected residents from lujiang county with high gastric cancer risk in Eastern China.* Biol. Trace Elem. Res., 186(1): 21-30. doi: 10.1007/s12011-018-1283-4

Hua X., Duan F., Huang J., Bi X., Xia W., Song C., Wang L., Jiang C., Yuan Z. 2021. *A novel prognostic model based on the serum ıron level for patients with early-stage triple-negative breast cancer.* Front Cell Develop Biol, 9: 777215. https://doi.org/10.3389/fcell.2021.777215

International Agency for Research on Cancer. *Global Cancer Observatory. 2021.* World Health Organization. Available at: https://gco.iarc.fr/ (date of access: 07.06.2021)

Jaafari-Ashkavandi Z., Khademi B., Malekzadeh M., Shahmoradi Z. 2019. *Serum levels of zinc, copper and ferritin in patients with salivary gland tumors*. Asian Pac. J. Cancer Prev., 26;20(2): 545-548. DOI: 10.31557/APJCP.2019.20.2.545 PMID: 30803218; PMCID: PMC6897009

Kaba M., Pirinççi N., Yuksel M.B., Gecit I., Gunes M., Ozveren H., Eren H., Demir H. 2014. *Serum levels of trace elements in patients with prostate cancer*. Asian Pac. J. Cancer Prev., 15(6):2625-2629.

Keshani F., Khalesi S., Aghaz A., Farhang M., Akbari N. 2021. *Screening of oral squamous cell carcinoma by serum changes: A systematic review and meta-analysis*. Dental Res J, 18: 88.

Khan N., Afridi HI., Kazi T.G., Arain M.B., Bilal M., Akhtar A., Khan M. 2017. *Correlation of cadmium and magnesium in the blood and serum samples of smokers and non-smokers chronic leukemia patients*. Biol. Trace Elem. Res., 176(1): 81-88. DOI: 10.1007/s12011-016--0816-y

Khanna S.S., Karjodkar F.R. 2006. *Circulating immune complexes and trace elements (copper, ıron and selenium) as markers in oral precancer and cancer : a randomised, controlled clinical trial*. Head Face Med, 2: 33. https://doi.org/10.1186/1746-160X-2-33

Khanna S., Udas A.C., Kumar G.K., Suvarna S., Karjodkar F.R. 2013. *Trace elements (copper, zinc, selenium and molybdenum) as markers in oral sub mucous fibrosis and oral squamous cell carcinoma*. J. Trace Elem. Med. Biol., 27(4): 307-11. DOI: 10.1016/j.jtemb.2013.04.003

Kriegel A.M., Soliman,A.S., Zhang Q., El-Ghawalby N., Ezzat F., Soultan A., Abdel-Wahab M., Fathy O., Ebidi G., Bassiouni N., Hamilton S.R., Abbruzzese J.L., Lacey M.R., Blake D.A. 2006. *Serum cadmium levels in pancreatic cancer patients from the East Nile Delta region of Egypt*. Environ Health Persp, 114(1): 113-119. https://doi.org/10.1289/ehp.8035

Lee M.H., Gao Y.T., Huang Y.H., McGee E.E., Lam T., Wang B., Shen M.C., Rashid A., Pfeiffer R.M., Hsing A.W., Koshiol J. 2020. *A metallomic approach to assess associations of serum metal levels with gallstones and gallbladder cancer*. Hepatology (Baltimore, Md.), 71(3): 917-928. https://doi.org/10.1002/hep.30861

Li H., Li X., Liu J., Jin L., Yang F., Wang J., Wang O., Gao Y. 2017. *Correlation between serum lead and thyroid diseases: papillary thyroid carcinoma, nodular goiter, and thyroid adenoma*. Int. J. Environ. Health Res., 27(5): 409-419. DOI: 10.1080/09603123.2017.1373273

Lichtenstein P., Holm N.V., Verkasalo P.K., Iliadou A., Kaprio J., Koskenvuo M., Pukkala E., Skytthe A., Hemminki K. 2000. *Environmental and heritable factors in the causation of cancer-analyses of cohorts of twins from Sweden, Denmark, and Finland*. N. Engl. J. Med., 343(2): 78-85.

López F., Rodrigo J.P., Cardesa A., Triantafyllou A., Devaney K.O., Mendenhall W.M., Haigentz M. Jr., Strojan P., Pellitteri P.K., Bradford C.R., Shaha A.R., Hunt J.L., de Bree R., Takes R.P., Rinaldo A., Ferlito A. 2016. *Update on primary head and neck mucosal melanoma*. Head Neck, 38(1): 147-55. DOI: 10.1002/hed.23872

Lopez J., Ramchandani D., Vahdat L. 2019. *Copper depletion as a therapeutic strategy in cancer*. Met. Ions Life Sci., 14;19:/books/9783110527872/9783110527872-018/9783110527872--018.xml, DOI: 10.1515/9783110527872-018

Lubiński J., Jaworowska E., Derkacz R., Marciniak W., Białkowska K., Baszuk P., Scott R.J., Lubiński J.A. 2021. *Survival of laryngeal cancer patients depending on zinc serum level and oxidative stress genotypes*. Biomolecules, 11(6): 865. https://doi.org/10.3390/biom11060865

Massardier V., Catinon M., Trunfio-Sfarghiu A.M., Hubert J., Vincent, M. 2020. *Metal-metal hip prosthesis and kidney cancer: assumed role of chromium and cobalt overload*. Am J Case Rep, 21: e923416. https://doi.org/10.12659/AJCR.923416

Morais S., Costa F.G., Pereira M.D.L. 2012. *Heavy metals and human health*. Chapter 10. In: *Health-Emerging Issues and Practice*. Environ. J. Oosthuizen (Ed.).

Naja G.M., Volesky B. 2009. *Toxicity and sources of Pb, Cd, Hg, Cr, As, and radionuclides in the environment*. Chapter 2. In: *Handbook on Heavy Metals in the Environment*.

Neethi H., Patil S., Rao R.S. 2013. *Estimation of serum copper and zinc levels in oral submucous fibrosis: an atomic absorption spectroscopic study*. J. Contemp. Dent. Pract., 1;14(5): 801-5. DOI: 10.5005/jp-journals-10024-1406 PMID: 24685778

Nevitt T., Ohrvik H., Thiele D.J. 2012. *Charting the travels of copper in eukaryotes from yeast to mammals*. Biochim. Biophys. Acta, 1823. 1580-1593.

Okunade K.S., Dawodu O.O., Salako O., Osanyin G.E., Okunowo A.A., Anorlu R.I. 2018. *Comparative analysis of serum trace element levels in women with invasive cervical cancer in Lagos, Nigeria*. Pan. Afr. Med. J., 20;31: 194. DOI: 10.11604/pamj.2018.31.194.14425

Olson A. D., Hamlin W. B. 1969. *A new method for serum ıron and total ıron-binding cabacity by A.A.S.* Clin. Chem., 15(6): 438-44.

Olszewski J., Latusiński J., Kita A., Pietkiewicz P., Starostecka B., Majak J. 2006. *Comparative assessment of aluminum and lead concentrations in serum and tissue bioptates in patients with laryngeal papilloma or cancer*. B-ENT, 2(2): 47-9. PMID: 16910286

Ozmen H., Erulas AF., Karatas F., Cukurovali A., Yalcin O. 2006. *Comparison of the concentration of trace metals (Ni, Zn, Co, Cu and Se), Fe, vitamins A, C and E, and lipid peroxidation in patients with prostate*. Clin. Chem. Lab. Med., 44: 175-179.

Pirinççi N., Gecit I., Gunes M., Kaba M., Tanik S., Yuksel MB., Arslan H., Demir H., 2013. *Levels of serum trace elements in renal cell carcinoma cases*. A. Pac. J. Cancer Prev, 14: 499-502.

Polter E.J., Onyeaghala G., Lutsey P.L., Folsom A.R., Joshu C.E., Platz E.A., Prizment A.E. 2019. *Prospective association of serum and dietary magnesium with colorectal cancer ıncidence*. Cancer Epidemiol Biomarkers Prev., 28(8): 1292-1299. DOI: 10.1158/1055-9965. EPI-18-1300

Rajizadeh A., Mozaffari-Khosravi H., Zavar-Reza J., Shiryazdi S.M. 2017. *Comparison of hematological parameters, iron levels, and oxidative stress in women with and without breast cancer: A case – control study*. Med J Islamic Rep Iran, 31: 114. https://doi.org/10.14196/ /mjiri.31.114

Rashed M.N. 2006. *Fruit stones from industrial waste for the removal of lead ions from polluted water*. Environ Monit Assess, 119: 31-41.

Reis A.M.D., Fruchtenicht A.V.G., Athaydes L.C., Loss S., Moreira L.F. 2017. *Biomarkers as predictors of mortality in critically ill patients with solid tumors*. An. Acad. Bras. Cienc., 89(4): 2921-2929. DOI: 10.1590/0001-3765201720170601

Rhee J.,Graubard B.I., Purdue M.P. 2021. *Blood lead levels and lung cancer mortality: An updated analysis of NHANES II and III*. Cancer Med, 10(12): 4066-4074. https://doi. org/10.1002/cam4.3943

Risby T.H. 2006. *Modern ınstrumental analysis*. In: *Comprehensive Analytical Chemistry*. Ahuja S., Jespersen N. (Eds.). Vol. 47, Chapter 8.

Rzymski P., Tomczyk K., Poniedziałek B., Opala T., Wilczak M. 2015. *Impact of heavy metals on the female reproductive system*. Ann Agric Environ Med., 22(2): 259-64. DOI: 10.5604/ /12321966.1152077. PMID: 26094520

Saleh S., Adly H.M., Abdelkhaliq A.A., Nassir A.M. 2020. *Serum levels of selenium, zinc, copper, manganese, and ıron in prostate cancer patients*. Curr. Urol., 14(1): 44-49. https://doi.org/ /10.1159/000499261

Satarug S., Garrett S.H., Sens M.A., Sens D.A., 2010. *Cadmium, environmental exposure, and health outcomes*. Environ Health Persp, 118(2): 182-190.

Shanbhag V.C., Gudekar N., Jasmer K., Papageorgiou C., Singh K., Petris, M.J. 2021. *Copper metabolism as a unique vulnerability in cancer*. Biochim Biophys Acta Mol Cell Res., 1868(2): 118893. https://doi.org/10.1016/j.bbamcr.2020.118893

Song X., Zhong X., Tang K., Wu G., Jiang Y. 2018. *Serum magnesium levels and lung cancer risk: a meta-analysis*. World J. Surg. Oncol., 12;16(1): 137. DOI: 10.1186/s12957-018-1447-x

Sukiennicki G.M., Marciniak W., Muszyńska M., Baszuk P., Gupta S., Białkowska K., Jaworska--Bieniek K., Durda K., Lener M., Pietrzak S., Gromowski T., Prajzendanc K., Łukomska A., Waloszczyk P., Wójcik J. Z., Scott R., Lubiński J., Jakubowska A. 2019. *Iron levels, genes involved in iron metabolism and antioxidative processes and lung cancer incidence.* PloS One, 14(1): e0208610. https://doi.org/10.1371/journal.pone.0208610

Taysi S., Akcay F., Uslu C., Dogru Y., Gulcin I. 2003. *Trace elements and some extracellular antioxidant protein levels in serum of patients with laryngeal cancer.* Biol Trace Elem Res., 91(1): 11-18. DOI: 10.1385/BTER:91:1:11 PMID: 12713025

Tian T., Xiao F., Li H., Ding D., Dong W., Hou G., Zhao L., Yang Y., Yang Y., Zhou, W. 2021. *Association between serum iron status and primary liver cancer risk: a Mendelian randomization analysis.* Ann Transl Med, 9(20): 1533. https://doi.org/10.21037/atm-21-4608

Tiwari R., David C.M., Mahesh D.R., Sambargi U., Rashmi K.J., Benakanal P. 2016. *Assessment of serum copper, iron and immune complexes in potentially malignant disorders and oral cancer.* Braz Oral Res., 10;30(1):e101. DOI: 10.1590/1807-3107BOR-2016.vol30.0101 PMID: 27737355

Türkdoğan M.K., Karapinar H.S., Kilicel F. 2022. *Serum trace element levels of gastrointestinal cancer patients in an endemic upper gastrointestinal cancer region.* J Trace Elem Med Biol., 25;72: 126978. DOI: 10.1016/j.jtemb.2022.126978

Wang Y., Sun Z., Li A., Zhang Y. 2019. *Association between serum zinc levels and lung cancer: a meta-analysis of observational studies.* World J Surgical Oncol, 17(1): 78. https://doi.org/10.1186/s12957-019-1617-5

Woźniak A., Kujawa A., Seńczuk-Przybyłowska M., Kulza M., Gawecki W., Szybiak B., Herman M., Czarnywojtek A., Kurhańska-Flisykowska A., Chesy P., Szyfter W., Walas S., Golusiński W., Szyfter K., Krejpcio Z., Piekoszewski W., Parczewski A., Florek E. 2012. *Physiological metals in the serum, hair and nails of patients with head and neck cancer.* Prz. Lek. 69(10): 785-97. (in Polish). PMID: 23421034

Xie Y., Wang J., Zhao X., Zhou X., Nie X., Li C., Huang F., Yuan H. 2018. *Higher serum zinc levels may reduce the risk of cervical cancer in Asian women: A meta-analysis.* J Int Med Res, 46(12): 4898-4906. https://doi.org/10.1177/0300060518805600

Yaman M., Kaya G., Simsek M. 2007. *Comparision of trace elements concentrations in cancerous and noncancerous human endometrial and ovary tissues.* Int J. Gynecol Cancer, 17: 220-228.

Yıldız İ. 2015. *Serum trace element levels in the patients with bladder cancer.* (Master thesis). T.C Yüzüncü Yıl University, İnstitute of Naturel and Applied Sciences, Department of Chemistry Van, Turkey. (in Turkish)

Zabłocka-Słowińska K., Płaczkowska S., Prescha A., Pawełczyk K., Porębska I., Kosacka M., Pawlik-Sobecka L., Grajeta H. 2018. *Serum and whole blood Zn, Cu and Mn profiles and their relation to redox status in lung cancer patients.* J Trace Elem Med Biol., 45: 78-84. DOI: 10.1016/j.jtemb.2017.09.024

Zhang X., Yang Q. 2018. *Association between serum copper levels and lung cancer risk: A meta-analysis.* J Int Med Res, 46(12): 4863-4873. https://doi.org/10.1177/0300060518798507

Zhang M., Shi M., Zhao Y. 2018. *Association between serum copper levels and cervical cancer risk: a meta-analysis.* Biosci Reports, 38(4): BSR20180161. https://doi.org/10.1042/BSR20180161

Zanoni D.K., Montero P.H., Migliacci J.C., Shah J.P., Wong R.J., Ganly I., Patel S.G. 2019. *Survival outcomes after treatment of cancer of the oral cavity (1985-2015).* Oral Oncol, 90: 115-121. https://doi.org/10.1016/j.oraloncology.2019.02.001