Ann. For. Res. 65(1): 2371-2385, 2022
ISSN: 18448135, 20652445

ANNALS OF FOREST RESEARCH
https://www.e-afr.org/

Evaluation of certain serum trace element levels in patients with acute and chronic leukemia

Omar H. A. Abuhmid[1], Sukayna H. Rashed[2]
[1,2] Department of Chemistry sciences, College of Science, University of Mosul, Mosul, Iraq
[2]sukaynarashed@uomosul.edu.iq [1]abuhamidoh2@gmail.com

ABSTRACT
The study's major focus was on how trace elements were distributed in blood samples from patients with leukemia.The current study focuses on assessing potential connections between trace elements and acute and chronic kinds of leukemia through the measurement of their amounts in blood serum. In patients, Cd was found at sub-ppm levels, with Mg being the main contributor at mean levels. The mineral copper plays a crucial role in controlling the body's metabolism, which is necessary for a variety of biological processes. Generally speaking, an increased risk of malignancies, particularly blood cancers, is associated with iron deficiency. It can be crucial if the body's level of it changes noticeably. The current study measured the amounts of copper, magnesium, iron, and cadmium in the sera of leukemia patients who had been referred to Mosul Hospitals and compared the results with a control group's findings. Evaluation included 30 healthy people and 60 leukemia patients. Patients were divided into groups with acute and chronic leukemia. Both the control group and the patients' blood was drawn. using the flame atomic absorption technique, copper and cadimium levels in the sera were determined, while magnesium and iron were measured coloremetrically. 31 (51.66%) men and 29 (48.33%) women made up the patient population, whereas 15 (50%) men and 15 (50%) women made up the healthy participants. The average age of those in the patient group was 22.5 years, whereas that of those in the control group was 23.9 years. The age ranges for the case group and the control group were 3-71 years and 2-79 years, respectively. Acute leukemia (30 instances) and chronic leukemia were among the patients in the cohort (30 cases). Serum copper levels in patients were noticeably higher ($p< 0.001$) when compared to controls. An enormous rise in the levels of copper, cadmium, and iron, and a slight and insignificant decrease in the rates of magnesium concentrations in leukemia patients compared with healthy subjects. Evidence of elevated levels of Cu, Cd and Fe was observed in leukemia patients, which could imply that these substances play a function in cancer as a distinctive risk factor for malignancy.

KEYWORDS: Leukemia, Copper, Cadimium, Iron, Magnesium

INTRODUCTION
Leukemia is a sort of cancer that can affect anyone, regardless of age, and refers to a wide range of ailments with vastly different biological bases, such as malignancies of hematopoietic cells (1). Leukemia, often known as blood cancer, comes in both acute and chronic forms, with several major kinds (2). It also has a huge costs of diagnosis and treatment and affects people of all ages (3) Blood cancer cases, like leukemia, have been rising throughout human societies (4). A variety of clinical and pathological signs are used to diagnose leukemia. Numerous environmental

elements have been linked to various disorders from the perspective of their origin (5-11). The development of neoplastic disease is influenced by a variety of internal and environmental factors, including the existence of trace elements. Trace elements have been the subject of in-depth research, trace elements contribute significantly to human health (12The biological effects of trace elements can be described by their ability to promote the start of free radical reactions as pro-oxidants or anti-oxidants, or by promoting the breakdown of peroxides and other unstable molecules (13). Therefore, it is crucial to study the distribution and function of different metals in relation to specific disorders. Copper is a crucial mineral in the control of the body's immunity among all minerals (14), Cu is necessary to maintain the body's blood vessels, epithelium and connective tissues, and skin's strength. It keeps the thyroid glands working normally and is crucial for the creation of hemoglobin, myelin, and melanin. In the blood, ceryloplasmin, which transports copper to tissues, contains around 90% of the copper (15, 16). All living things require copper as a necessary cofactor and antioxidant (17). Cu is an antioxidant that can lessen some of the harm that free radicals produce by scavenging or neutralizing them (18, 19). Because it is an oxidant, it might encourage the damage caused by free radicals, which might help with Alzheimer's (20). Blood copper levels have been reported to change in some malignant tumors as lung, breast and gastrointestinal malignancies (21). Over 30 enzymes require copper as a cofactor (22). eukaryotic cells' cytoplasm contains the antioxidant enzyme superoxide dismutase, which has copper at its catalytic site (21). Additionally, catecholamine and ATP-producing enzymes' chemical structures both depend on copper (21). Copper is a component of growth factors, including the endothelium growth factor, which, in response to a rise in serum copper levels, promotes tumor progression through angiogenesis (23). According to earlier studies, the rise in copper levels caused a rise in lipid peroxidation, the breakdown of the antioxidant system, and the production of radicals, consequently, the DNA gets harmed, which causes cancer-causing mutations (24). The majority of magnesium is contained in soft tissue—both muscular and non-muscular (25). It vitally stabilizes enzymes, including several ATP-generating activities, serves as a cofactor in more than 300 enzyme processes (26, 27). If magnesium metabolism is disturbed, it may have an impact on ATP-dependent processes (28). Magnesium inhibits the calcium-dependent release of acetylcholine at motor endplates, making it a natural "calcium antagonist" (29, 30).

Fe performs essential bodily tasks , but a Fe overload can result in acute Fe poisoning. Chronic poisoning may also result from excessive Fe ingestion or repeated blood transfusions (13). Anemia in general and iron deficiency anemia (IDA) are associated with a higher risk of several malignancies, notably blood cancers. The blood levels of hazardous divalent cations, particularly cadmium (Cd^{2+}), are known to be higher in both IDA subjects and smokers. An established carcinogen is cadmium. The majority of the circulating cadmium is linked to transferrin, and in addition to the kidney and liver, which are the target organs for cadmium accumulation, tissues (cells) with high levels of transferrin receptor 1 (TfR1) expression may also collect large amounts of circulating cadmium. In bone marrow cells, the density of TfR1, a glycoprotein expressed on cell surfaces, is not constant. In the bone marrow, megakaryocyte/erythrocyte progenitors and pro-erythroblasts express TfR1 considerably more than other cell lines. We believe that these cell types

will take up the majority of the circulating cadmium and afterwards be the most appropriate for malignant transformation. A toxic metal (Cd) can replace a necessary element, disrupting crucial metabolic processes. Toxic elements in particular compete with essential ones in the synthesis of ligands with enzymes and other proteins (31). Thus, even brief and low-concentration exposure to hazardous metals may result in epigenetic changes and mutations that later result in various cancers. Leukemia has been linked to essential element overloads or deficits in a number of studies, with both acute (32, 33, 39) and chronic (40) leukemia being positively correlated with Cu (32, 40) and Fe (36, 37, 41) levels. To our knowledge, however, only a small number of studies have been conducted regarding the association between hazardous trace element levels in serum and leukemia (31-41), suggesting substantial associations between high Cd levels and acute (42, 37) or chronic (42, 37) leukemia (36). Recent years have seen a significant increase in research on the role of trace elements in biological processes, specifically variations in copper, iron, magnesium, and cadimium concentrations and their connection to malignancy (43, 44). As a result, the current study sought to compare the serum levels of copper, iron, magnesium, and cadimium in acute and chronic leukemia patients with those of healthy donors. The study investigates any relationship between them and the likelihood of developing both acute and chronic leukemia kinds.

MATHERIALS AND METHODS

The study, which had 60 leukemia patients (acute and chronic leukemia) hospitalized at AL-SALAM and IBN AL-ATHEER Hospitals in Mosul, Iraq, ran from December 2021 to July 202230 healthy people were chosen and given a thorough medical exam in order to compare the results. The ethics committee at the University of Mosul in Iraq's Mosul provided the inspiration for the study's design. Each participant in the study gave their voluntary consent in writing and signed it. Clinical exams (biochemical, hematological, microbiological, serological, and hormone testing) were used to validate the group's normal health state and revealed no signs of leukemia or other disorders. Clinical testing supported the leukemia diagnosis made for the patient populationPatients and the control group who took vitamin and mineral supplements were not included in the study. A total of 60 case samples and 30 control samples were examined. Gel tubes with fasting blood samples were filled, and the tubes underwent a 10-minute centrifugation at 3000 rpm. After being separated from the clots, sera was kept at -20°C. By using an atomic absorption with flame technique, the total content of copper and cadimium in sera was determined (Perkin Elmer Aanalyst 100; Perkin Elmer). Using (5M) HNO3, samples were diluted. Utilizing several standards' dilutions, the standard curves were produced. The amounts of copper and cadimium were measured in comparison to a standard curve. Utilize the ready-made analysis kit from the Italian manufacturer (LTA) to calculate the magnesium level in blood serum. Using the colorimetric approach, ready-made solutions from the French business BioLabo were used to estimate the amount of iron in the blood serum.

STATISTICAL ANALYSIS

SPSS, or the Statistical Package for the Social Sciences, was used to examine the data (version 21.0, Nie, Bent, and Hull, USA). Mean and standard deviation were used to display descriptive

data. Paired t-test was used to analyze the serum levels of trace components. Statistics were deemed to be significant at P-values 0.05.

RESULTS

The information of leukemia patients and the age- and healthy controls is shown in **Table 1**. The healthy participants consisted of 15 (50%) males and 15 (50%) females, while the sick were made up of 31 (51.66%) males and 29 (48.33%) females. The average age of those in the patient group was 22.5 years, whereas that of those in the control group was 23.9 years. The age ranges for the case group and the control group were 3-71 years and 2-79 years, respectively. Both acute (30 cases) and chronic (30 cases) leukemia patients made up the patient population (**Table 1**).

Table 1 patient and control group characteristics

patients and normal group	n	M (%)	F (%)
Chronic leukemia	30	16 (53.33)	14 (46.66)
Acute leukemia	30	15 (50)	15 (50)
Total	60	31 (51.66)	29 (48.33)
Control group	30	15 (50)	15 (50)

As shown in **Table 2**, the average serum copper concentration for patients was 151.6952.1 µg/dL and 94.9351.2 µg/dL for healthy people, showing a significant difference (p =0.015).

Table 2 Mean trace element concentrations in patients' and a control group's sera

Trace elements	control group n = 30 (mean ± standard deviation)	patients group n = 60 (mean ± standard deviation)	P value
Cu (µg/dL)	94.935±1.2	151.695±2.1*	0.015
Cd (µg/dL)	0.951± 0.04	2.717±0.12*	0.035
Fe (µg/dL)	49.892± 1.02	63.238± 1.14*	0.044
Mg (mg/dL)	2.78± 0.64	1.54±0.41	0.212

The mark * indicates that the groups differ significantly from one another at the level of probability ($p \leq 0.05$).

n = sample count

Also, there were significant increase in Cd and Fe levels in patients group as compared with control (p=0.035 and 0.044) for Cd and Fe respectively.

When studying the effect of the duration of leukemia on the trace elements levels of patients with leukemia for the group of total patients, **Table (3)** patients with a period of more than a year and patients with a period of less than a year) do not vary considerably from one another. And for both sexes for the male group and the female group **Tables (4) and (5)**, respectively.

Table 3 Effect of the duration of leukemia on the levels of trace elements in blood serum for the total group of patients

Trace elements	The duration of leukemia		P- value
	More than a year n = 31 (mean ± standard deviation)	Less than a year n = 29 (mean ± standard deviation)	
Cu (µg/dL)	4.5 ± 151.18	2.6 ± 142.63	0.77
Cd (µg/dL)	0.15 ± 3.03	0.16 ± 2.51	0.073
Fe (µg/dL)	7.6 ± 81.68	1.6 ± 61.53	0. 073
Mg (mg/dL)	0.05 ± 1.63	0.35 ± 1.52	0.77

The mark * indicates that the groups differ significantly from one another at the level of probability ($p \leq 0.05$).

Table 4 Effect of duration of leukemia on the levels of studied elements in the blood serum of a group of male patients

Trace elements	The duration of leukemia		P- value
	More than a year n = 18 (mean ± standard deviation)	Less than a year n = 13 (mean ± standard deviation)	
Cu (mg/dL)	150.04± 10.1	158.6± 7.1	0.77
Cd (mg/dL)	2.59± 0.08	2.64± 0.1	0.67
Fe (mg/dL)	68.6± 2.1	58.82± 7.1	0.53
Mg (mg/dL)	4.10 ± 1.9	7.10 ± 1.6	0.57

The mark * indicates that the groups differ significantly from one another at the level of probability ($p \leq 0.05$).

Table 5 Effect of leukemia duration on levels of trace elements in a group of female patients' blood serum

Trace elements	The duration of leukemia		Pvalue
	More than a year n = 13 (mean ± standard deviation)	Less than a year n = 16 (mean ± standard deviation)	
Cu (mg/dL)	154.65± 9.2	146.65± 7.9	0.7
Cd (mg/dL)	2.15± 0.04	2.05± 0.09	0.73
Fe (mg/dL)	76.65± 5.9	66.65± 1.29	0.73
Mg (mg/dL)	1.65± 0.08	1.55± 0.09	0.509

The mark * indicates that the groups differ significantly from one another at the level of probability ($p \leq 0.05$).

There were no effect of leukemia type on elements levels in patients with leukemia, for both sexes Table (6).

Table 6 Effect of leukemia type on levels of trace elements in blood serum

Trace elements	The type of leukemia		Pvalue
	acute n = 30 (mean ± standard deviation)	chronic n = 30 (mean ± standard deviation)	
Cu (mg/dL)	154.65± 9.2	146.65± 7.9	0.7
Cd (mg/dL)	2.15± 0.04	2.05± 0.09	0.73
Fe (mg/dL)	76.65± 5.9	66.65± 1.29	0.73
Mg (mg/dL)	1.65± 0.08	1.55± 0.09	0.509

The mark * indicates that the groups differ significantly from one another at the level of probability ($p \leq 0.05$).

When studying the effect of age on copper levels for the group of male leukemia patients and the group of female leukemia patients, **Table (7) and (8)**, respectively, found an increase in copper levels in the third age group compared to the younger groups and for both sexes.

Table 7 In male patients, the age-related effects on trace elements

Trace elements	Male patients n = 31		
	Age groups (years)		
	Less than 15 (mean ± standard deviation) n=9	40) – 15((mean ± standard deviation)n=11	greater than 40 (mean ± standard deviation)n=11
Cu (µg / dL)	145.81±0.8 a	149.31± 0.116 a	151.62± 7.11 b
Cd (µg / dL)	2.305± 0.03 a	2.511± 0.06 a	2.05± 0.09 a
Fe (µg / dL)	52.635±0.16 a	64.075± 0.8 b	64.15± 0.1 b
Mg (µg / dL)	1.045± 0.09 a	1.26± 0.27 c	1.211± 0.02 b

The different letters in the horizontal position denote a statistically significant difference ($p \leq 0.05$) between the groups.

Table 8 age effect on trace elements of female patients

Trace elements	Female patients n = 29		
	Age groups (year)		
	Less than 15)mean±SD(n=12	40) – 15( )mean±SD(n=6	Over 40)mean±SD(n=11
Cu (µg / dL)	147.6±0.4 a	150.23±4.4 b	0.04 ± 152.57 c
Cd (µg / dL)	2.31±0.03 a	2.29±0.15 a	0.017 ± 2.54 a
Fe (µg / dL)	52.44±0.07 a	52.44±0.005 a	64.903±0.09 b
Mg (µg / dL)	1.095±0.07 a	1.265±0.1 b	1.13±0.02 a

The different letters in the horizontal position denote a statistically significant difference ($p \leq 0.05$) between the groups.

The results indicated that there was no effect of gender on copper levels in patients with leukemia, for both sexes ,Table (9).

Table 9 Gender differences in trace element levels between the control and leukemia patient groups

Trace elements	Control (n=30)		Patients (n=60)	
	Male (n=15) mean±SD	Female(n=15) mean±SD	Male (n=15) mean±SD	Female(n=15) mean±SD
Cu (µg / dL)	97.32±0.3 a	89.433±0.1 a	148.85±2.6 b	150.13±1.6 b
Cd (µg / dL)	0.848±0.06 a	1.02±0.03 b	2.35±0.06 c	2.38±0.06 c
Fe (µg / dL)	44.4±0.06 a	43.42± 0.8 a	60.19±0.3 b	56.59±0.05 b
Mg (µg / dL)	2.448±0.1 b	2.19± 0.06 a	1.17±0.1 a	1.163±0.08 a

A statistically significant difference between the groups is indicated by the various letters in the horizontal position ($p \leq 0.05$).

The results in **Table (6)** indicate that there is no effect of cancer type on cadmium rates for acute leukemia patients group with chronic leukemia group for both sexes.

Also, there was no effect of sex on cadmium levels in the group of leukemia patients, Table (9) The results in **Tables (3), (4) and (5)** indicate that there are no significant differences when studying the effect of the duration of leukemia on the iron levels of the total group of patients, the group of male patients, and the group of female patients, respectively.

When studying the effect of the leukemia kind on iron levels, the findings indicated in **Table (6)** that there was a significant increase in the rate of iron for chronic leukemia patients compared to acute leukemia patients in the male group. When studying the effect of age on iron levels, the results in **Table (7)** indicated that iron levels were higher for the second and third age groups, compared with the younger age group in the male group. This may be due to the more severe oxidative stress in the second and third groups, while **Table (8)** indicates a rise in iron levels for the older age group compared to the two younger groups in the female group. The reason may be due to the increase in oxidative stress, the effect of estrogen and menopause. There is also no effect of gender on iron levels for leukemia patients,**Table (9).** When studying the effect of age on magnesium levels for leukemia patients, the results in **Tables (7) and (8)** indicate that there are more magnesium levels in the second age group (15-40) years compared to the younger and older age groups.

DISCUSSION

Trace elements, which make up less than 0.01% of the body's weight, are essential for fundamental cellular processes. With the established roles, it was discovered that changed blood levels of the major trace elements were linked to various prenatal disorders, including ALL. Not

only does chemotherapy damage one's nutritional state, but it also depletes macro- and micronutrients (45). Any large variations in the level of this element could be hazardous to the body due to copper's nutritional importance, crucial functions in metabolism control (46) and direct connection with malignancies (21), among other factors (47). According to numerous studies, an increase in copper causes the antioxidant system to be destroyed, increased MDA causes the generation of radicals, which assaults DNA and results in cancer-causing mutations, to grow (24). Meanwhile, the endothelium growth factor, is thought to include copper, which is a part of the chemical makeup of enzymes that make ATP as well as catecholamine hormones, when serum copper levels rise, the endothelial growth factor promotes tumor development and angiogenesis (23). A lot of research has been done on the element Cu in cancer. Patients with lymphoma had a greater amount of it discovered, but after treatment, there was a noticeable drop and the serum level stayed within the normal range during remission. Cu levels rising was thought to be a sign of relapse. Regarding leukemia, children who were in remission were found to have normal levels of Cu, and it was discovered that the active form of the disease altered this element (48). Our findings supported the earlier finding that Cu levels considerably dropped during the first chemotherapy induction. Age, gender, and treatment protocol had little impact on the element.

Despite the fact that Alkufi et al. (2015) found a large increase in Cu serum levels among ALL patients, a comparison research between these patients and healthy people revealed lower levels of Cu among ALL patients (49). Similarly, following the commencement of chemotherapy, our patients experienced copper shortage. Copper levels in the study's leukemia patients were higher than those in the control group ($p < 0.001$). The findings of the current investigation were consistent with those of (50), who found that individuals with acute myeloid leukemia had greater serum copper levels than did healthy subjects. (44) revealed that, in comparison to healthy people, copper levels were significantly higher in leukemia patients. In line with the findings of this investigation, Carpentieri et al. (1986) found no association between gender and copper levels in the normal or sick groups (51). Additionally, serum copper levels were not substantially correlated with age, supporting the findings of (52). One of the least affected elements was magnesium, which had no reports of insufficiency either before or after the start of chemotherapy. Compared to our findings, Demir et al. observed increased levels of this element following therapy. Along with what Afridi et al. had previously reported, they also noticed that patients with ALL had reduced Mg serum levels. They proposed that Mg may be utilized as a predictive index in patients with ALL in response to chemotherapy by linking the greater level of Mg after chemotherapy with the release of this element from damaged cells (53). Therefore, it is possible that the abnormally high quantities of those same trace metals in the current study's leukemia patients will have an effect on how the disease develops. Iron has been demonstrated to have a carcinogenic effect in vivo and to have a dual role in malignancies (54). Patients with acute leukemia who were also susceptible to fungal infections had elevated serum iron levels (55). 14% of ALL patients reported having iron excess by (56). Some doctors utilize iron chelators as part of their treatment in relation to the role of iron that has been discussed (57). Patients who were treated under high risk protocols and those who were older than 10 years old lost more iron following chemotherapy, which may be related to

the low rate of blood transfusion among our patients. By causing cell mutations, elevated iron levels in patients' blood may also raise their risk of developing cancer (58). Here, it was discovered that serum levels of toxic Cd, which has numerous negative health effects (59), were substantially greater in patients than controls. Its interaction with erythrocytes, which affects blood shape and hemoglobin production (60), and its high dose induction of leukocytosis and anemia in humans (61) are likely the causes of the detected increased levels. Additionally, The encouragement of trace elements entering the circulation may be due to the metals' relatively long retention times in the bones—up to 30 years for Cd's half-life (62). Natural killer cells may also be negatively impacted by toxic trace elements (63), and greater Cd concentrations have been connected to a rise in non-Lymphoma Hodgkin's (64)and leukemia in mice following Cd exposure (65). Other investigations have suggested that long-term Cd exposure may cause bone marrow cells and peripheral blood lymphocytes to become genotoxic and cytotoxic (66, 67). Ohanian et al(68) .'s recent study, which looked at AML patients' survival, showed that lower survival was seen in cases with greater harmful trace element levels. Similar outcomes were also seen when vital trace metals like Cu or Fe exceeded a specific threshold and turned poisonous (68). These findings are consistent with those of the current investigation. With the exception of hazardous Cd, which tends to bioaccumulate (62, 66) Most of the examined trace elements were identified in increased concentrations in the serum of people with acute forms of leukemia, and is also involved in various epigenetic processes in cancer (67). The K562 cell's line downregulated DNA owing to Cd induction was investigated in Huang's et al(69) study on Cd in CML, indicating an increase in cell proliferation as a result of the presence of Cd.

CONCLUSION

This study evaluated the serum copper, cadimium, and iron levels in leukemia patients. Elevated concentrations of studied trace elements were found in patients of leukemia, which could indicate that the element has a role in cancer and may serve as a separate risk factor for malignancy.

REFERENCES

I. Polychronakis, G. Dounias, V. Makropoulos, E. Riza, A. Linos, Work-related leukemia: a systematic review, J. Occup. Med. Toxicol. 8 (2013), https://doi.org/10.1186/1745-6673-8-14, 1.

2- D.M. Bozzone, The Biology of Cancer: Leukemia, Infobase, New York, 2009.

3- Fauci A., Braunwald E., kasper D., Hauser S., longo D. and jameson J. (2008). Harrison's principles of internal medicine. 17th ed. MncGraw- Hill, New York. 687 -690.

4- Brewer J.G. (2001). Copper control as an antiangiogenic anticancer therapy: Lessons from Treating Wilson S Disease. *Exp. Biol. Med.* 226: 665- 73.

5- M. Belson, B. Kingsley, A. Holmes, Risk factors for acute leukemia in children: a review, Environ. Health Perspect. 115 (2007) 138–145, https://doi.org/10.1007/s00411005007510.1289/ehp.9023.

6- J.E. Heck, J. Wu, C. Lombardi, J. Qiu, T.J. Meyers, M. Wilhelm, M. Cockburn, B. Ritz, Childhood cancer and traffic-related air pollution exposure in pregnancy and early life, Environ. Health Perspect. 121 (2013) 1385–1391, https://doi.org/ 10.1289/ehp.1306761.
7- N. Winters, M.S. Goldberg, P. Hystad, P.J. Villeneuve, K.C. Johnson, Exposure to ambient air pollution in Canada and the risk of adult leukemia, Sci. Total Environ. 526 (2015) 153–176, ttps://doi.org/10.1016/j.scitotenv.2015.03.149.
8- P. Fern´andez-Navarro, J. García-P´erez, R. Ramis, E. Boldo, G. L´opez-Abente, Industrial pollution and cancer in Spain: an important public health issue, Environ. Res. 159 (2017) (2017) 555–563, https://doi.org/10.1016/j. envres.2017.08.049.
9- D. Xu, D. Liang, Y. Guo, Y. Sun, Endosulfan causes the alterations of DNA damage response through ATM-p53 signaling pathway in human leukemia cells, Environ. Pollut. 238 (2018) 1048–1055, https://doi.org/10.1016/j.envpol.2018.03.044.
10- C. Lin, Y. Hsu, K.D. Brown, B. Pokharel, Y. Wei, S. Chen, Residential exposure to petrochemical industrial complexes and the risk of leukemia: a systematic review and exposure-response meta-analysis, Environ. Pollut. 258 (2020), 113476, https://doi.org/10.1016/j.envpol.2019.113476.
11- K.G. Koukoulakis, P.G. Kanellopoulos, E. Chrysochou, V. Koukoulas, M. Minaidis, G. Maropoulos, G.P. Nikoleli, E. Bakeas, Leukemia and PAHs levels in human blood serum: preliminary results from an adult cohort in Greece, Atmos. Pollut. Res. 11 (2020) 1552–1565, https://doi.org/10.1016/j.apr.2020.06.018.
12- Schubert HL, Wilson KS, Raux ESC, Woodcock SC, Warren MJ (1998) The X-ray structure of a cobalamin biosynthetic enzyme, Coprecorrin- 4 methyltransferase. Nat Struc Biol 5: 585-592.
13- Fraga CG, Oteiza PI (2002) Iron toxicity and antioxidant nutrients, Toxicology 180: 23–32.
14-Bilbis L.S., Idowu D.B., Saidu Y., Lawal M. and Njoku C.H. (2010). Serum levels of antioxidant Vitamins and mineral elements of human immunodeficiency virus positive subjects in Sokoto. *Nigeria. Ann Afr Med.* 9: 235-9.
15- Harris ED (2001) Copper homeostasis: the role of cellular transporters, Nutr Rev 59: 281-285.
16- Rottkamp CA, Nunomura A, Raina AK, Sayre LM, Perry G, et al. (2000) Oxidative stress, antioxidants, and Alzheimer's disease. Alzheimer Disease Assoc Disorders 14: 62-66.
17- Chen Y.W., Chen K.L., Chen C.H., Wu H.C., Su C.C.and Wu C.C. (2010). Pyrrolidine dithiocarbamate (PDTC)/Cu complex induces lung epithelial cell apoptosis through Mitochondria and ER-stress pathways. *Toxicol. Lett.* 15: 333-40.
18- Araya M, Pizarro F, Olivares M, Arredondo M, Gonzalez M, et al. (2006) Understanding copper homeostasis in humans and copper effects on health, Biol Res 39: 183-187.
19- Bonham M, Jacqueline M, Bernadette MH, Strain JJ (2002) The immune system as a physiological indicator of marginal copper status? Brit J Nut 87: 393-403.
20- Davis CD (2003) Low dietary copper increases fecal free radical production, fecal water alkaline phosphatase activity and cytotoxicity in healthy men. J Nutr 133: 522-527.

21- Zarghami N., Asadi J., Mahbob S., Mohammadzadeh G. and Mohajeri A. (2008). Serum levels of Se, Zn, Cu and Cu / Zn Ratio in Iranian Breast Cancer Patients. *Pharmaceutical Sci.* Spring: 27- 32.
22- Silvia L., Anna M. and Alessandra M. (2005). Mechanism– based in activators of plant copper/ quinone containing amine oxidases. Photochem. 7: 1751- 58.
23- Kim S.Y., Kim J.W., koo J.E., Chung H.Y. and Lee –Kim Y.C. (2003). Changes in lipid eroxidation and antioxidant trace elements in serum of women with cervical intraepithelial neoplasia and invasive cancer . *Nutr. Cancer.* 47 (2): 126- 30.
24- Wu T., Sempos C.T., Muti P. and Smit E. (2004). Serum Iron, Copper and Zinc concentrations and risk of cancer mortality in US adults. *Ann. Epidamiol.* 14 (3): 195- 201.
25- Das KK, Gupta AD, Dhundasi SA, Patil AM, Das SN, et al. (2006) Effect of L-ascorbic acid on nickel-induced alterations in serum lipid profiles and liver histopathology of rats . J Basic Clin Physiol Pharmacol 17: 29-44.
26- Elin RJ (2010) Assessment of magnesium status for diagnosis and therapy. Magnes Res 23: 194-198.
27- Saris NE, Mervaala E, Karppanen H, Khawaja JA, Lewenstam A (2000) An update on physiological, clinical and analytical aspects. Clin Chim Acta 294: 1-26.
28- Aikawa JK (1981) Magnesium: Its Biological Significance. Boca Raton, FL: CRC Press.
29- Geiger H, Wanner C (2012) Magnesium in disease. Clin Kidney J 5: i25-i38.
30- Wacker W (1980) Magnesium and Man. Cambridge, MA: Havard University Press 1-184.
31- M. Abdulla, J. Chmielnicka, New aspects on the distribution and metabolism of essential trace elements after dietary exposure to toxic metals, Biol. Trace Elem. Res. 23 (1989) 25–53, https://doi.org/10.1007/BF02917176.
32- X.L. Zuo, J.M. Chen, X. Zhou, X.Z. Li, G.Y. Mei, Levels of selenium, zinc, copper, and antioxidant enzyme activity in patients with leukemia, Biol. Trace Elem. Res. 114 (2006) 41–53, https://doi.org/10.1385/BTER:114:1:41.
33- O.R. Zekavat, M. Karimi, F. Majidi, M. Bordbar, S. Haghpanah, S. Parand, H. Bozorgi, Trace elements in children with acute lymphoblastic leukemia, Asian Pac. J. Cancer Prev. 21 (2020) 43–47, https://doi.org/10.31557/APJCP.2021.22. S1.43.
34- A. Modaressi, M. Hadjibabaie, A.R. Shamshiri, R. Namdar, M. Abdollahi, A. Ghavamzadeh, Trace elements (Se, Zn, and Cu) levels in patients with newly diagnosed acute leukemia, Int. J. Hematol. Stem. Cell. Res. 6 (2012) 5–10.
34- Swaminathan R (2003) Magnesium metabolism and its disorders. Clin Biochem Rev 24: 47-66.
35- E.A.A. Elshaygi, Assessment of serum levels of electrolytes and trace elements in leukaemia patients in Sudan, Acta Sci. Pharm. Sci. 2 (10) (2018) 136–141.
36- C.G.L. Canellas, S.M.F. Carvalho, E.F.O. De Jesus, M.J. Anjos, R.T. Lopes, Trace and major elements in serum of patients with chronic myelogenous leukemia, J. Radioanal. Nucl. Chem. 269 (2006) 631–634, https://doi.org/10.1007/ s10967-006-0276-5.

37- C. Demir, H. Demir, R. Esen, A. Sehitogullari, M. Atmaca, M. Alay, Altered serum levels of elements in acute leukemia cases in Turkey, Asian Pac. J. Cancer Prev. 12 (2011) 3471–3474.
38- H.K. Alkufi, Determination the levels of zinc and copper in patients with leukemia, Int. J. Curr. Microbiol. App. Sci. 4 (2015) 812–816.
39- S. Valadbeigi, S. Javadian, M. Ebrahimi-Rad, S. Khatami, R. Saghiri, Assessment of trace elements in serum of acute lymphoblastic and myeloid leukemia patients, Exp. Oncol. 41 (1) (2019) 69–71.
40- M. Gundogdu, H. Kaya, I. Gulcin, F. Erdem, K. Cayir, M. Keles, A. Yilmaz, Oxidase Activity of Ceruloplasmin and Some Acute Phase Reactant and Trace Elements Concentrations in Serum of Patients With Chronic Lymphocytic Leukemia, 52(1), 2007, p. 24.
41- P. Fenaux, C.J.Br. Rose, Impact of iron overload in myelodysplastic syndromes, Blood Rev. 23 (2009) 15–19.
42- J. García-P´erez, G. L´opez-Abente, D. G´omez-Barroso, A. Morales-Piga, E. P. Romaguera, I. Tamayo, P. Fern´andez-Navarro, R. Ramis, Childhood leukemia and residential proximity to industrial and urban sites, Environ. Res. 140 (2015) 542–553, https://doi.org/10.1016/j.envres.2015.05.014.
43- Navarro S.A. and Rohan T.E. (2007). Trace elements and cancer risk: review of the epidemiologic evidence. *Cancer Causes Control.* 18(1): 572- 8.
44- Zarghami N., Alizadeh F., Ansarin K.H. and Mohajerr A. (2009). Correlation between serum levels of zinc and copper and teleomerase gene expression in lung cancer patients. *Pharmaceutical Sci.* 14 (4): 183- 190.
45- Modaressi A, Hadjibabaie M, Shamshiri A, et al (2015). Trace elements (Se, Zn, and Cu) levels in patients with newly diagnosed acute leukemia. *Int J Hematol Oncol Stem Cell Res*, 6, 5-10.
46- Fukai T. and Ushio-Fukai M. (2011). Superoxide Dismutases: Role in Redox Signaling, Vascular Function and Diseases. Antioxid Redox Signal. 15 (6): 1583- 1606
47- Jing L., Wu Y., Wu J., Zhao J., Zuo D. and Peng S. (2011). Peroxiredoxins are involved in metallothionein protection from doxorubicin cardiotoxicity. *Eur. J. Pharmacol.* 659 (2–3): 224–232.
48- Federico A, Iodice P, Federico P, et al (2001). Effects of selenium and zinc supplementation on nutritional status in patients with cancer of digestive tract. *Eur J Clin Nutr*, 55, 293.
49- Alkufi HK (2015). Determination the levels of Zinc and Copper in patients with leukemia. *Int J Curr Microbiol Appl Sci*, 4, 812-6.
50-Qunzhi H., Jiexian J., Xianwen Z., Jingang G., Suling H. and Gozdasoglu S. (2003). Classification and prognostic value of serum Copper/ Zinc ratio in Hodgkin's disease. *Biol. Trace. Elem. Res.* 91(2): 191- 2.
51- Carpentieri U., Myers J., Thorpe L., Daeschner C.W. and Haggard M.E. (1986). Copper, zinc, and iron in normal and leukemic lymphocytes from children. *Cancer Res.* 46(2): 981- 4.
52- Sgarbieri B., Fisberg M. and Tone L.G. (1999). Nutritional assessment and serum zinc and copper concentration in leukemic children. *Sao Paulo Med. J.* 117(1): 13-18.

53- Afridi HI, Kazi TG, Talpur FN (2018). Correlation of Calcium and Magnesium levels in the biological samples of different types of acute leukemia children. *Biol Trace Elem Res*, 86, 395-406.
54- Toyokuni S (2009). Role of iron in carcinogenesis: cancer as a ferrotoxic disease. *Cancer Sci*, 100, 9-16.
55- Fenaux P, Rose CJBr (2009). Impact of iron overload in myelodysplastic syndromes. *Blood Rev*, 23, 15-9.
56- Halonen P, Mattila J, Suominen P, et al (2003). Iron overload in children who are treated for acute lymphoblastic leukemia estimated by liver siderosis and serum iron parameters. *Pediatrics*, 111, 91-6.
57- Estrov Z, Tawa A, Wang X-H, et al (1987). In vitro and in vivo effects of deferoxamine in neonatal acute leukemia. *Blood*, 69, 757-61.
58- Reddy SB, Charles MJ, Raju GJN, Reddy BS, Reddy TS et al. (2004) Trace elemental analysis of cancer-afflicted intestine by PIXE technique. Biol Trace Elem Res 102: 265-281.
59- M. Jaishankar, T. Tseten, N. Anbalagan, B.B. Mathew, K.N. Beeregowda, Toxicity, mechanism and health effects of some heavy metals, Interdiscip. Toxicol. 7 (2014) (2014) 60–72, https://doi.org/10.2478/intox-2014-0009.
60- Y. Dai, X. Huo, Y. Zhang, T. Yang, M. Li, X. Xu, Elevated lead levels and changes in blood morphology and erythrocyte CR1 in preschool children from an e-waste area, Sci. Total Environ. 592 (2017) 51–59, https://doi.org/10.1016/j.scitotenv.2017.03.080.
61- B.G. Luckett, L.J. Su, J.C. Rood, E.T.H. Fontham, Cadmium exposure and pancreatic cancer in South Louisiana, J. Environ. Public Health 2012 (2012), 180186, https://doi.org/10.1155/2012/180186.
62- W.J. Choi, S.H. Han, Blood cadmium is associated with osteoporosis in obese males but not in non-obese males: the Korea National Health and Nutrition Examination Survey 2008–2011, Int. J. Environ. Res. Public Health 12 (2015) 12144–12157, https://doi.org/10.3390/ijerph121012144.
63- Y. Zhang, X. Huo, J. Cao, T. Yang, L. Xu, X. Xu, Elevated lead levels and adverse effects on natural killer cells in children from an electronic waste recycling area, Environ. Pollut. 213 (2016) 143–150, https://doi.org/10.1016/j.envpol.2016.02.004.
64- B.R. Blakley, The effect of cadmium on chemical- and viral- induced tumor production in mice, J. Appl. Toxicol. 6 (1987) 425–429, https://doi.org/10.1002/jat.2550060608.
65- R.S. Kelly, T. Lundh, M. Porta, I.A. Bergdahl, D. Palli, A.S. Johansson, M. Botsivali, P. Vineis, R. Vermeulen, S.A. Kyrtopoulos, M. Chadeau-Hyam, Blood erythrocyte concentrations of cadmium and lead and the risk of B-cell non-Hodgkin's lymphoma and multiple myeloma: a nested case-control study, PLoS One 8 (2013), e81892, https://doi.org/10.1371/journal.pone.0081892.

66- J. Choiniere, L. Wang, Exposure to inorganic arsenic can lead to gut microbe perturbations and hepatocellular carcinoma, Acta Pharm. Sin. B 6 (2016) 426–429, https://doi.org/10.1016/j.apsb.2016.07.011.
67- S. Kim, J.H. Freeland-Graves, M. Babaei, P.K. Sachdev, S.N. Beretvas, Quantifying the association between acute leukemia and serum zinc, copper, and selenium: a meta-analysis, Leuk. Lymphoma 60 (2019) 1548–1556, https://doi.org/10.1080/10428194.2018.1540043.
68- M. Ohanian, P. Telouk, S. Kornblau, F. Albarede, P. Ruvolo, R.S.S. Tidwell, A. Plesa, R. Kanagal-Shamanna, E.L. Matera, J. Cortes, A. Carson, C. Dumontet, A heavy metal baseline score predicts outcome in acute myeloid leukemia, Am. J. Hematol. 95 (2020) 422–434, https://doi.org/10.1002/ajh.25731.
69- D. Huang, Y. Zhang, Y. Qi, C. Chen, W. Ji, Global DNA hypomethylation, rather than reactive oxygen species (ROS), a potential facilitator of cadmium-stimulated K562 cell proliferation, Toxicol. Lett. 179 (2008) 43–47, https://doi.org/10.1016/j.toxlet.2008.03.018.