The association between serum level of zinc and copper and prevalence of bladder cancer in Iraq

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Abstract

Bladder cancer (BC) is the most common malignant tumor in the urinary tract and the tenth most common malignant tumor throughout the world, with an estimated 4:1 male-to-female ratio. Men are primarily affected by it. Bladder cancer is the fourth most common malignant tumor in men and the eighth most common cancer in women in Iraq. Copper is an essential component of cells. Copper can participate in a variety of reactions by acting as an electron donor or recipient copper could have a role in the etiology, severity, and progression of cancer disease. Zinc the second most common trace element in the human body. Zinc plays a critical role in healthy human physiology and has also been linked to a few pathophysiological conditions. This study aimed to assess the level of zinc and copper in bladder cancer patients as well as the impact of changes in these minerals on the disease. Cu in patients' group was significantly higher (146.09 ± 14.06 µg/dl) in compared to control (109.42 ± 17.04 µg/dl), p-≤ 0.001. while the mean of Zn in patients' group was significantly lower (68.87 ± 11.13mg/dl) than mean in control group (98.20 ± 14.33mg/dl), p-≤ 0.001. The mean of Zn/Cu ratio in patients' group was significantly lower (0.48 ± 0.11) than mean in control group (0.92 ± 0.22), p ≤ 0.001.

Keywords: Bladder Cancer, Zn and Cu ratios, Serum, Copper, Zinc.

1. Introduction

Bladder cancer (BC) is the most common prevalent malignant tumor in the

urinary tract and the tenth most common malignant tumor worldwide [1]. Malepredominant and predicted to have a 4:1 male-to-female ratio, approximately 570,000 individuals were diagnosed with bladder cancer in 2020, making it one of the top 10 most prevalent malignant tumors worldwide [2]. Bladder cancer ranks fourth among malignant tumors in men and eighth among cancers in women in Iraq. Bladder cancer can strike people at an early age, with over 90 % of newly diagnosed cases occurring in those over 55 [3].

There are two main types of bladder cancer primary and secondary. Primary bladder cancer is those that start in the bladder itself. Secondary bladder cancers are those that originated in another organ and then spread to the bladder. Other tumors can transfer to the bladder through the lymphatic system, bloodstream. or by directly separating from a closed organ, such as the prostate or the rectal. Cancers beginning in the bladder are more common than cancers that metastasis to the bladder from another location. Urothelial CA Bladder classified into four group transitional cell carcinoma (TCC), squamous cell carcinoma (SCC), adenocarcinoma and mixed carcinoma [4].

Urologic surgeons, or urologists, use the term progression to describe how likely it is for a cancer to grow and spread or to recur. Frequently, they base treatment plans on the stage, World Health Organization (WHO)- International Society of Urologic Pathology Consensus Classification, using the following categories: non-invasive bladder cancer and muscle invasive bladder cancer [5]. Tobacco smoking is the biggest risk factor for bladder cancer. Environmental or occupational pollutants also significantly increase the disease burden [6].

Bladder cancer is also associated with advanced age and male sex [7]. Bladder tumors may show up as microscopic or gross hematuria, which is assessed using upper tract imaging and cystoscopy based on the severity of the hematuria and the likelihood of malignancy [8]. An essential component of cells, copper can participate in a variety of reactions by acting as an electron donor or recipient. However, high concentration of copper ion can cause free radicals and oxidative stress in cells [9]. Copper could have a role in the etiology, severity, and progression of cancer disease [10].

The second most common trace element in the human body is zinc (Zn). Based on its vital roles in bone metabolism, growth and development, the central nervous system, immunological function, and wound healing. Zinc is significant for human health and disease [11]. Zinc plays a critical role in healthy human physiology and has also been linked to a high number of pathophysiological conditions, such as depression, cancer, diabetes mellitus, heart disease, and neurodegenerative diseases like Alzheimer's [12].

As one of the most common trace metal imbalances, zinc/copper ratio is clinically more important than the concentration of any other trace metals. Zinc/copper ratio is an index associated with the imbalance of oxidative stress. Disruption of Cu-Zn homeostasis may lead to oxidative stress, a state where increased formation of reactive oxygen species overwhelms body antioxidant protection and subsequently induces DNA lipid peroxidation, damage, protein modification and possibly cancer development [13].

2. Methodology

This study case-control study was conducted at the University of Baghdad, College of Medicine, Department of Biochemistry from January to June of 2023. Patients were selected from Ghazi Hariri Hospital in Bagdad. Moreover, ninety people were recruited in the current study and divided into two groups.

Forty-five patients were selected for the first group that included 37 males, and 8 females with an age range between 47 and 82 years. However, thirty-five patients are smokers for a period of 5 years or longer. The second group also included 45 healthy individuals 37 males, and 8 females with normal bladder tissue and without any previous history of any other renal systemic diseases.

All patients attended the hospital and diagnosed with primary superficial bladder cancer and exclusion patients with metastatic tumor other types of malignancy renal disease malnutrition. Each subject had approximately 10 milliliters of venous blood that were drawn between 9:00 am and 1:00 pm. The blood was then emptied into gel tubes, allowed to clot, and centrifuged for 10 minutes at 3000 rpm to extract serum. The serum was then transferred into Eppendorf tubes with labels and frozen at -80 °C until the zinc and copper levels were determined.

Furthermore, collected serum samples were frozen and left to melt at room temperature. Copper and zinc inorganic elements were evaluated using flame atomic absorption spectrometry (FAAS) as shown in (figure 1). Moreover, (figure 1) illustrates the calibration curve that was created by measuring the absorbance of standard solutions at various concentrations and plotting the concentration on a graph. After the absorbance was determined, the unknown

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element's concentration in the sample was removed [14].

Working calibration solutions of copper and zinc were prepared from stock solutions. The prepared stock solutions were from zero concentration, to at three concentrations or more to set the base line and read zero absorbance. Blank solutions concentration standards) (zero were solution aspirated. Moreover, blank preparation step was repeated multiple times to correct the base line drift. The work standard solutions were examined in a stepwise manner and started with the most diluted solution to the most concentrated [15]. The normal range of copper in the serum is 62 μ g/dl to 140 μ g/dl (14 to 42.49 mmol/l) where copper was measured at wavelength 324 nm [16]. On the other hand, the normal ranges of zinc in the serum are 80 to 120 mg/dL (12 to 18 mmol/L) where zinc was measured at wavelength 214.86 nm [17].

3. Results

The current study included 90 participants that were divided into two groups equally. Regarding gender, 16 females (8 patients and 8 healthy control) and 74 males (37 patients and 37 healthy control), with no significant association between studied group and gender, p-value 1.00 [18].



Figure 1: distribution age of patients.

Concerning age group samples, 5.6 % were below 40 years, 20 % were 41-50 years. While 34.4 % were 51-60 years, 27.8 % were 61-70 years, and 12.2 % were above 70 years. A significant association between older age group (> 70 years) and being a patient and a significant association between lower age group (< 40 years) and healthy control, pvalue 0.008. The mean of age in patients' group $(61.47 \pm$ 11.28 years); was significantly greater than the control group's mean. $(58.87 \pm 9.41 \text{ years}); p \le 0.03 \text{ as shown}$ in (table 1).

		Groups			n	
Infor	mation	Patients	Contr ol	Total	value	
		8	8	16		
Contra	Male	17.8 %	17.8 %	17.8 %	1.00	
Gender	Female	37	37	74	1.00	
		82.2 %	82.2 %	82.2 %		
	D 1 40	0	5	5		
	years	0.0 %	11.1 %	5.6 %		
	41-50 years	10	8	18		
		22.2 %	17.8 %	20.0 %		
Age	51-60 years	12	19	31	0.008	
groups		26.7 %	42.2 %	34.4 %	*	
	61-70 years	13	12	25		
		28.9 %	26.7 %	27.8 %		
	Older than	10	1	11		
	70 years	22.2 %	2.2 %	12.2 %		
Age (mean ± SD)		61.47 ± 11.28	$\begin{array}{c} 58.87 \\ \pm 9.41 \end{array}$		0.03*	
Smoking	Nonsmokers	12	40	52		
		26.7 %	88.9 %	57.8 %	<	
	Smokers	33	5	38	*	
		73.3 %	11.1 %	42.2 %		
		45	45	90		
Total		100.0 %	100.0 %	100.0 %		

Table 1: Association between demographicdata and studied groups.

*p-value is significant

The mean \pm SD of copper serum (Cu) in patients' groups (146.09 \pm 14.06 µg/dl) significantly higher than the mean \pm SD of groups control (109.42 \pm 17.04 µg/dl), where p- \leq 0.001 as presented in (table 2). The mean \pm SD of zinc in patients' group was significantly lower (68.87 \pm 11.13 mg/dl) than mean \pm SD in control group (98.20 \pm 14.33 mg/dl), p- \leq 0.001 also presented in (table 2). The mean \pm SD of Cu/Zn ratio in patients' group was significantly lower (0.48 \pm 0.11) than mean \pm SD in control group (0.92 \pm 0.22), p \leq 0.001.

Table	2 : Mean	\pm SD o	of Cu,	Zn	and	Cu/Zn
ratios	in the stuc	lies grou	ıps.			

Trace Elements	Group	Mean ± SD	SE	p-value
Serum Zn mg/dl	Patients	68.87 ± 11.13	1.65	<
	Control	98.20 ± 14.33	2.13	0.001*†
Serum Cu µg/dl	Patients	146.09 ± 14.06	2.09	<
	Control	109.42 ± 17.04	2.54	0.001*†
Serum Zn/Cu	Patients	0.48 ± 0.11	0.01	*-
	Control	0.92 ± 0.22	0.03	

Zinc (Zn) has an inverse significant moderate correlation with copper (Cu). (r -0.56, p. ≤ 0.001), and copper (Cu) has inverse significant moderate correlation with the zinc (Zn) (r -0.56, p. ≤ 0.001) as presented in (table 3).

Table 3: Correlation of included parameter inboth groups.

Group	Zn	Cu	
Zn	/	- 0.56**	
Ln	/	< 0.001	
Cu	- 0.56**	1	
Cu	< 0.001	/	

4. Discussion

The results of the regarding age groups, listed in (table1) explain the mean age of studied population was older than 60 years. A high number of studies support our results for example research that was held at the University of Bagdad with the age of the studied population was 63.5 ± 11.4 years and the most prevalent age was more than 60 years [19]. In adult males ≥ 60 years urinary bladder cancer was the most common type and representing 11.94 % of cancers in other studies at the University of Basrah [20].

Other publication in Iran supported our study results as the age group over sixty is the most vulnerable to develop bladder cancer [21]. Based on (table1) results a significant association between smoking and patients' group (73.3 % vs 26.7 %) p- \leq 0.001. This is consistent with all global and local research, such as smoking increases the risk of tumors recurrence and progression. While it is still controversial whether smoking cessation in bladder cancer will favorably influence the outcome of bladder cancer treatment, patients should be counselled to stop smoking because of the general risks connected to tobacco smoking [22]. Also, a high cigarette smoking in Iran is linked to a higher risk of bladder cancer as opposed to never using smoking[23]. The benefit of copper is listed in (table 2) as increasing the percentage beyond the permissible limit causes serious adverse effects. An excess of copper may lead to other diseases in the body including cancer. These results of the present study agree with results of other study conducted in Iraq for example, patients' group's serum copper level was statistically significantly higher than the control group's ($p \le 0.001$) [14].

Another research in Egypt concluded that plasma levels of Cu were measured in 60 matched controls and 66 patients with transitional bladder cell carcinoma (BC group) [24]. The results showed that plasma Cu were significantly higher in bladder cancer (BC) group when compared with the control group [24]. Since Cu ions may be involved in the activation of signaling pathways related to cell proliferation, the role of Cu in the progression of cancer has long been a subject of study. Research indicates that when compared to healthy, quiescent cells, cancer cells typically have a higher demand for copper. Furthermore, reports of elevated Cu concentrations in tumor tissues and/or serum from patients suffering from a variety of cancers, including bladder cancer, have been made. Also, in patients with bladder cancer, a high serum of Cu levels has been linked to the advancement of the disease as well as the stage of the tumor [25].

Zinc plays an important role in preventing cancer as shown in (table 2). Zn also plays an important role in reducing malignancy as an antioxidant and a component of many DNA repair proteins. Zinc plays a crucial function in protecting DNA from damage as antioxidant, antiinflammatory and proapoptotic. These results of the present study agree with results of other studies conducted that conclude the zinc serum level in patients' group was significantly lower than in control group (p - ≤ 0.001) [14]. Moreover, the bladder cancer (BC) group's plasma zinc (Zn) level was much lower than the controls. When the levels of Cu and Zn in the bladder tissues of the bladder cancer (BC) group were compared with those in the non-cancerous bladder tissues [24]. Results showed that the cancerous tissues had significantly higher Cu levels and significantly lower Zn levels. Finally changes in the plasma and bladder tissue concentrations of Cu and Zn are associated with the etiology of bladder cancer [24].

5. Conclusion

The current study concludes that smoking is one of the main causes of bladder cancer disease. The increase in copper concentration in patients with bladder cancer, as well as a decrease in the percentage of zinc concentration in this disease. Nutritional supplements can be used to fill this deficiency. There was also a decrease in the ratio of zinc to copper compared to the control. Therefore, the researcher notes that there is an inverse relationship between an increase in copper concentration and a decrease in copper concentration in patients with bladder cancer in the Bagdad city.

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