



## Estimation of Serum Copper, Zinc and Iron in Patients with Oral Cancer

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### ABSTRACT

The aim of the study is to estimate serum copper, zinc and iron in patients with oral cancer. Higher incidence of oral cancer in Indian population necessitates in depth probing of various etiological and contributing factors for its early diagnosis and prognosis. Trace elements are versatile anti cancer agents that regulate various biological mechanism. Apart from the crucial role of micronutrients like copper, iron, and zinc in the functions of body enzymes, it seems that changes in the serum levels of these biomarkers may play a role in the pathogenesis of oral cancer. To evaluate the serum levels of copper, zinc and iron in oral cancer patients and to suggest that these trace elements could be used as bio marker in oral cancer. Serum level of copper, zinc and iron in patients with oral cancer was estimated.

**Keywords:** copper, zinc, iron, serum, trace elements.

### INTRODUCTION

India has one of the highest incidence of oral cancer in the world. The development of cancer is a multi step process arising from pre existing potentially malignant lesion. Leukopenia is the most common precancer representing 85% of such lesion. Alcohol consumption, Genetic mechanisms and chronic irritation have modifying effects in the etiology of oral cancer. The occurrence of oral cancer has been rising during the last decade in some countries and it is the sixth fatality cause in the world, as approximately 50% of diagnosed patients die annually with this disease<sup>1</sup>. The burden of cancer is increasing in economically developing countries as a result of growth and aging population. The adoption of a lifestyle with high risk habits due to rapid economic development further compounds the impact of cancer. Worldwide, oral cancer is ranked as the eleventh most common type of cancer with 130,000 reported deaths annually. It is the most commonly seen in south and south East Asian countries such as India, Bangladesh, Taiwan and Srilanka<sup>2</sup>. Increasing incidence of oral cancer and precancer in India population necessitates in depth probing of various etiological and contributory factors for its early diagnosis and prognosis. Recently trace elements are receiving much attention in the detection of oral cancer and precancerous as they found to be significantly altered in the neck, lung and breast carcinomas. Trace elements have an important role in carcinogenesis<sup>3</sup>. Trace elements have been regarded as versatile anti cancer agents that regulate various biological mechanisms. Trace elements are also required in small concentrations as essential components of biological enzymes systems or structural portions of biologically active constituents. Many metabolic disorders, oral precancerous conditions and oral cancer are accompanied by alterations in the concentrations of one or more trace elements like

copper, zinc, magnesium, iron in some body fluids, especially blood serum or plasma. Serum trace elements levels also helps to counteract the oxidative stress induced by free radicals which can cause serious damage to cells in oral precancerous conditions and oral cancer can help not only in the early diagnosis and treatment but also in prognosis as the disease progresses<sup>4</sup>. Zinc and copper have been the most extensively studied of the trace elements in patients with malignant disease and these elements in serum has been found to be reliable parameter as a diagnostic and prognostic index in case of craniofacial tumours<sup>5</sup>.

Zinc is one of the antioxidant in food on which the activities of enzymes of the antioxidant system of the body such as superoxide dismutase. Copper is an essential trace element required for the functioning of several key enzymes like cytochrome c oxidase, superoxide dismutase, metallathionein and Lysol oxidase. Zinc is implicated in modulation of mucosal metallathionein interfering with copper absorption<sup>6</sup>.

Iron has been recognised as an important element for maturation of epithelium and it is well documented that iron deficiency is associated with epithelial abnormalities in addition to malignancies like post cirroid carcinoma and tumours of pharynx and mouth<sup>7</sup>. Iron is one of the main micronutrient whose insufficiency that is prevalent all over the world may cause oral cancer by inducing oxidative events and ability to remove them by enzymatic and enzymatic antioxidants results in oxidative stress. This reaction can cause serious damage to cells and DNA because of producing excessive free radicals or decreasing the antioxidant defence defence or both. Oxidative damage are mostly followed by decline in theft cation of antioxidant enzymes.

Decrease or increase rare element such as Zinc, Copper, and Iron may influence the activity of antioxidant enzymes as well. The objective of this study was to measure the concentrations of serum copper, iron and zinc in patients with oral cancer and compare it with control groups.

**MATERIALS AND METHODS**

This cross sectional study was conducted on 15 patients diagnosed with oral cancer and 15 controls. Subjects of the control group neither had cancer nor suffered from systemic disease.

Controls were matched with cancer group in terms of age and gender. In patients of cancer institute were examined after obtaining required authorization.

Then a data form comprising demographic and characteristics of lesions, as well as informed written consent were completed for them.

**Collection of Blood Samples**

At the hospital, 5ml of blood sample were collected from the patients intravenously by registered nurses and then transferred to acid -wash tubes.

Then the samples were centrifuged, the serum part was separated and kept in a refrigerator at 70 degrees, finally the samples were checked for minimising error.

**RESULTS**

**Estimation of Serum Copper**

Serum copper was estimated by colorimetric method using a procedure mentioned in the kit manufactured by coral clinical systems.

**Estimation of Serum Zinc**

Serum zinc was estimated by nitro papas method using AGappe kit. The absorbance of standard and test was measured against the blank using colorimeter method.

**Estimation of Serum Iron**

Serum iron was estimated using AGappe kit by ferrozine method. The absorbance of standard and test was measured against the blank.

**Statistics**

Mean and standard deviation of the quantitative values of micronutrients in patients and healthy people were determined. Quantities of ferritin, copper and zinc in the two groups of patients and controls were compared using Mann- Whitney U non parametric test because these variables was not normal. Therefore, student’s t test was used for statistical analysis. Spss software version 18 was used for statistical analysis.

**Table 1: NPAR Test**

Group Statistics					
	Group	N	Mean	Std. Deviation	Std. Error Mean
Iron	Test	15	66.27	21.262	5.490
	Control	15	151.20	49.160	12.693
Zinc	Test	15	235.87	87.105	22.490
	Control	15	115.87	42.572	10.992
Copper	Test	15	315.00	302.809	78.185
	Control	15	233.60	229.462	59.247

Independent Samples Test

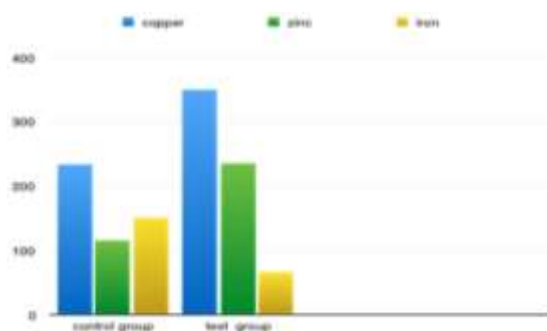
		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Iron	Equal variances assumed	3.539	.070	-6.141	28	.000	-84.933	13.829	-113.262	-56.605
	Equal variances not assumed			-6.141	19.061	.000	-84.933	13.829	-113.875	-55.994
Zinc	Equal variances assumed	3.179	.085	4.794	28	.000	120.000	25.033	68.722	171.278
	Equal variances not assumed			4.794	20.327	.000	120.000	25.033	67.836	172.164
Copper	Equal variances assumed	.533	.471	1.197	28	.241	117.400	98.007	-83.543	318.343
	Equal variances not assumed			1.197	26.091	.242	117.400	98.007	-84.207	319.007

**Table 2: Mann Whitney Test**

Ranks				
	Group	N	Mean Rank	Sum of Ranks
Copper	Test	15	20.43	306.50
	Control	15	10.57	158.50
	Total	30		



This graph explains the comparison of serum levels of copper, zinc and magnesium among control and test group.



**Graph 1**

This research was conducted on 15 patients with oral cancer and 15 healthy persons. Mean value of iron in the cancer group (66.27) micrograms/dl was significantly lower than in the control group (151.20) micrograms/dl. The mean concentration of copper (351.00) micrograms/dl in the cancer group were significantly higher than in the control group (233.60) micrograms/dl. Similarly mean concentration of zinc in the cancer group (235.87) were significantly higher than in the control group (115.87) micrograms/dl. So it is evident that serum values of copper and zinc in patients with oral cancer were high and iron levels were low when compared with the control group.

## DISCUSSION

Maryam baharvand studied that copper chelator compounds may act as tumor restraining factors. According to a research conducted in this regard, tetrathiomolybdate (TTM) has been found as a copper chelator in different kinds of cancer, which is capable of stopping tumor growth and preventing recurrence<sup>7</sup>. Theophanides and noted that copper ion plays a prominent role in producing free oxygen metabolites due to oxidation and regeneration activity<sup>8</sup>. Anastassopoulou and noted that zinc directly prevents DNA damage and eventually gene mutation and in this way, the element decreases the risk of cancer accordingly<sup>9</sup>. Sonali s Khanna and noted that serum iron levels are considered as biochemical indicators for nutritional assessment<sup>10</sup>. Utilisation of iron in collagen synthesis by the hydroxylation of proline and lysine leads to decrease serum iron level in patients with oral cancer. Reduction in the serum iron level may be due to malnutrition caused by tumours burden in cancer patients<sup>11</sup>. Jayadeep and et al noted that the rise in serum copper can be attributed to increased turnover of ceruloplasmin a copper carrying globulin with essential oxidase activity in the serum of carcinoma patients<sup>12</sup>. Vela D Desai and studied that the vital role played by micronutrients is revealed by the diversity of metabolic processes they help to regulate. On the contrary, unregulated and disproportionate level of transition metal ions (Fe<sup>2+</sup>, Cu<sup>2+</sup>) can catalyse and generate hydroxyl radicals (OH) from superoxide and

hydrogen peroxide via the Fenton reaction<sup>13</sup>. Banas A and noted that highly reactive hydroxyl radicals further cause lipid per oxidation and degradation of macromolecules, leading to cell damage or death<sup>14</sup>. Sham Kishor and studied that reduction in salivary trace elements in oral cancer patients may be explained on the basis that tumor cells and tissue have increased uptake from adjacent structures such as glandular secretion<sup>15</sup>. Al Rawi and the higher levels trace elements in saliva of oral cancer patients may be due to sequestration of these trace elements from cancer tissue to oral cavity, which is bathed by saliva<sup>16</sup>. Jyothi Tadakamadla and studied that among trace elements copper and iron are required for the functioning of numerous enzymes and therefore it is reasonable to assume that variation in serum level of these biochemical markers may be associated with the pathogenesis of oral cancer and precancerous states<sup>17</sup>. Nayak AG studied that lack of iron in the tissues results in decreased vascularity which facilitates percolation of arecoline<sup>18</sup>. Khanna S studied that trace elements play, directly or indirectly an important role in various physiological metabolic processes in human<sup>19</sup>. From the present study it was evident that serum levels of copper, zinc were increased in patients with oral cancer and iron levels were decreased in patients with oral cancer which correlates with findings.

## CONCLUSION

Serum levels of zinc and copper were higher in patients with oral cancer compared with controls. But serum level of iron in test group were lower than the control group. The present study it can be concluded as follows:

- ❖ It can be suggested that assessment of oral precancer and cancer patients may help in earlier diagnosis and/or prognosis.
- ❖ Utilization of iron in collagen synthesis by the hydroxylation of proline and lysine leads to decrease serum iron levels in patients with oral cancer.
- ❖ The rise in serum copper can be attributed to increased turnover of Ceruloplasmin in the serum of carcinoma patients.
- ❖ Anti fibrin and anti carcinogenic effects of zinc open the gateway to further research for supplementation of zinc for prevention of oral submucous fibrosis and its conversion to cancer.
- ❖ Trace metals affect each other's pharmacodynamics and pharmacokinetics.
- ❖ Also trace elements in correct proportion and concentration are essential for normal homeostasis.

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