The Relation between Trace Elements Levels and Some Cardiovascular Risk Factors in Patients with Obstructive Coronary Artery Disease in Basra

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Abstract

Objective: This study was performed in order to compare the levels of serum zinc, copper and iron in obstructive CAD patients with non CAD patients in Basra and to address the relation of these elements with some cardiovascular risk factors like Diabetes and hypertension.

Subjects and Methods: In a retrospective study, we evaluated 200 patients who underwent coronary angiography at AL- Basra Cardiac Center at Al-Sader Teaching Hospital. They were separated into two groups: case (patients with obstructive CAD) and non CAD. About two milliliters of venous blood samples were taken for measuring zinc, copper and iron. For statistical analyses, chi-square test, Student’s t-test and the logistic regression were used.

Results: The demographic and baseline clinical characteristics were not statistically different between the groups in terms of age, sex and BMI. Serum concentrations of zinc (56.60 ± 11.68 vs. 103.23 ± 20.62 µg/dl, p=0.0001) was statistically lower in patient group, serum copper (171.27 ± 28.87 vs. 121.33 ± 28.52 µg/dl, p=0.0001) was significantly higher in CAD patients while serum iron (113.33 ± 24.15 vs. 118.73 ± 23.95 µg/dl, p=0.115) was insignificantly tended to be lower in CAD patients. In subgroups of CAD patients according to DM and HT they had a significant (p value<0.05) higher level of copper and low level of zinc in diabetic and hypertensive than non-diabetic and normotensive respectively (p value<0.05). There was no statistical significant difference in the serum level of iron observed in these sub groups (p>0.05).

Conclusions: Low level of zinc and high level of copper may play a role in the pathogenesis of CAD while no such relation for serum iron as well as in diabetic and hypertensive CAD sub grouping; we found that these risk factors are affect positively on copper, while negatively on zinc. Moreover, insignificant effect is observed on serum iron level.

Keywords: Zinc; Copper; Iron; Obstructive coronary artery disease; Coronary angiography; Diabetes; Hypertension

Introduction

The leading cause of morbidity and mortality in developed countries is Coronary artery disease (CAD) and is evolving as an epidemic in developing countries. Traditional risk factors for CAD development including hypertension, hyperlipidemia, diabetes mellitus, age, sex, obesity, positive family history and cigarette smoking [1] although there is a strong evidence that oxidative free radicals have a role in the development of degenerative diseases and one of them is CAD [2]. Over the past 40 years, studies on the roles of trace elements in health and disease have led to a good understanding of their mechanism of function and why they are essential to human's life [3]. Because these trace elements zinc, copper and iron are micronutrients with known antioxidants and/or oxidant activity it might be relevant to assess their role in the atherogenesis in CAD [4].

Atherosclerosis is the principal cause of CAD [4]. Along with the theory of oxidative stress, atherosclerosis is the consequence of the oxidative modification of low density lipoproteins (LDL) in the arterial wall by reactive oxygen species (ROS). Evidence suggests that traditional risk factors for atherosclerosis upsurge the risk of the production of free ROS, not only from the endothelial cells, but also from the smooth muscle cells and the adventitial cells [5].

Copper is an important trace element and is associated with a number of metalloproteins [6], it is a catalytic component of numerous enzymes and is also a structural component of other important proteins, such as: Cytochrome c oxidase [7], l-lysyl oxidase [6-8]. Ceruloplasmin (feroxidase I), feroxidase II and haephestin in the Entocyte [6,8]. Both intracellular and extracellular superoxide dismutases are copper- and zinc- containing enzymes, able to convert superoxide radicals to hydrogen peroxide, which can be subsequently removed by catalase and other antioxidant defenses. Free Cu (II) ion can interact with hydrogen peroxide (H$_2$O$_2$) leading to the formation of the deleterious hydroxyl radical via the Fenton reaction [9].

Zn$^{2+}$ is an important player in the bodies' redox metabolism and zinc deficiency causes an elevation of oxidative stress [10]. Zn$^{2+}$ itself is not an antioxidant, because it does not take part in redox reactions. It is considered a proantioxidant [11]. This term describes several
antioxidant effects that zinc has, without directly participating in redox biochemistry. It can act through stabilization of cell membranes and sulfhydryls or as a structural component of antioxidant enzymes, such as copper/zinc superoxide dismutase \[12,13\].

**Subjects and Methods**

This study was conducted as a case control study and worked at Basra Medical College in the Department of Biochemistry on ninety-four patients (who attended AL-Basra Cardiac Center at Al-Sader Teaching Hospital, seeking for coronary angiography) with age ranging from 37 to 86 years old in whom coronary angiography revealed that they have >50% stenosis in one or more coronary arteries. One hundred and six patients from the same center their age range from 29 to 80 years old consider to be a non CAD patients group with a normal or ≤ 50% stenosis in one or more coronary arteries in coronary angiogram.

We classify the patient group according to the presence or absence of diabetes (diabetes which was defined as a known history of diabetes mellitus, fasting blood glucose >7 mmol/L (126 mg/dL) \[19\] or treatment with insulin or oral hypoglycemic agents) and hypertension (which was defined as a systolic blood pressure above 140 mmHg, or diastolic blood pressure above 90 mmHg \[20\] or current use of antihypertensive medication). The medical history and the results of angiograms were taken through special questionnaire.

Approximately two milliliters of venous blood samples were obtained from both non CAD patients individuals and patients with obstructive CAD. The separated serum were used for the assessment of serum zinc, copper and iron by Colorimetric method. Statistical analyses was done by chi-square test, Student's t-test, and the logistic regression.

**Results**

The study population composed from 94 angiographically documented obstructive CAD patients (mean age (59.25 ± 10.73) years), 71.2% of them are males, compared with 106 of non CAD patients and there is no statistical difference between them and CAD patients (p>0.05) in terms of age, sex, BMI as shown in Table 1.

There was a significant statistical difference in smoking behavior between them (p value <0.05) with CAD patients being more smoker than the contrary group. There were significantly higher prevalence of diabetes mellitus and hypertension in patients than non CAD patients group (p value <0.05).

Table 2 shows the classification of CAD group regarding the present or absent of DM and HT. In which there were 54.2% and 59.5% of patients had DM and HT respectively. From Table 3, it is clearly shown that the serum level of serum zinc was significantly (p value <0.05) lower in CAD patients than non CAD ones, while the serum level of copper was significantly higher in patient than non CAD patients group (p value <0.05).

Although, serum level of iron for patients group was lower than that of non CAD ones but it did not reach to the level of significance (p value >0.05).

<table>
<thead>
<tr>
<th>Variables</th>
<th>Obstructive CAD N=94</th>
<th>Non CAD N=106</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td>59.25 ± 10.73</td>
<td>57.21 ± 10.63</td>
</tr>
<tr>
<td><strong>Sex</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>67(71.2)</td>
<td>74(69.8)</td>
</tr>
<tr>
<td>Female</td>
<td>27(28.7)</td>
<td>32(30.2)</td>
</tr>
<tr>
<td><strong>BMI (kg/m²) mean ± SD</strong></td>
<td>29.84 ± 4.10</td>
<td>28.94 ± 4.09</td>
</tr>
<tr>
<td><strong>Diabetes mellitus n(%)</strong></td>
<td>51(54.2)</td>
<td>22(20.8)*</td>
</tr>
<tr>
<td><strong>Hypertension n(%)</strong></td>
<td>56(59.5)</td>
<td>37(34.9)*</td>
</tr>
<tr>
<td><strong>Smoking status n(%)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Current Smoker</td>
<td>30(31.9)</td>
<td>19(17.9)*</td>
</tr>
<tr>
<td>Non smoker</td>
<td>64(68)</td>
<td>87(82.1)*</td>
</tr>
</tbody>
</table>

*P value <0.05 and consider statistically significant.

**Table 1**: Demographic and clinical characteristics of the study population.
In logistic regression analysis, serum copper and zinc were significantly associated with CAD (OR= 0.743, 95% CI= 0.648-0.838, P =0.0001) and (OR =1.094, 95% CI=1.067 -1.123, P =0.0001) respectively after adjustment for traditional risk factors in the study population (Table 4), while no such association was found regarding the other factors. It is clear that there was a statistical difference in the serum level of Copper and Zinc (p value<0.05) between diabetic and diabetic free CAD patients in which copper was high and zinc was low in diabetic than non-diabetic patients as shown in Table 5.

However, there was no significant statistical difference in the serum level of iron in the same group, p value >0.05.

It is evident that statistically significant serum level of zinc tend to be low and serum copper tend to be high in hypertensive CAD patients (p value <0.05 each). While, no such finding regarding serum iron (p value >0.05), As shown in Table 6.

**Discussion**

The CAD patients and non CAD patients were matched in terms of age, sex, BMI Table 1. From the data collected in this study, The history of diabetes, hypertension and smoking were significantly differed between CAD and non CAD patients, being more in patients group, this finding was close to other study done by Alissa et al. [21] while differ from a study done by Cebi et al. [22]. This high prevalence of traditional risk factor ensure their role in the developments of CAD. Serum zinc and Copper remain statically significant after adjustment for other risk factors with logistic regression Table 4.

It is clearly found that serum level of zinc was significantly lower in patients group than that of non-obstructive CAD group (Table 3). This finding was in accordance with other finding [23,24] while disagreed with the other study [25] who found that zinc was not associated with the prevalence of CAD and with Cebi et al. [22] who found that serum zinc is low in the non CAD patients group rather than the patient, this different results could be due to differences in the method of assessing zinc or to differences in the number of patients and non CAD patients between these studies.

Zinc deficiency may be due to low dietary intake or possibly due to the presence of other nutrient that form complexes with zinc and prevent its absorption like fiber and phytate in which the phosphate groups in inositol hexaphosphate can form strong and insoluble complexes with cations such as zinc [26].

Regarding diabetic CAD patients, they had lower serum zinc level than non-diabetic CAD patients (Table 5). This finding has been substantiated by Mumtaz et al. [27] and other studies [28-30] and in the contrary with a study done in Gaza that revealed a high level of zinc in DM patients [31]. The cause of decreased serum zinc levels in diabetes may be an increase in urinary loss. Hyperglycaemia has been postulated to interfere with the active transport of zinc back in to the tubular cells. Other possible causes may be disturbed metabolisms of zinc metalloenzymes and an abnormal binding of zinc to tissue proteins, which cause low zinc level in DM [32].

It has been stated that hypertensive patients had lower zinc level than normotensive ones, this result is in accordance with [33,34] while disagreed with other [35]. Increased gastrointestinal absorption and urinary excretion of zinc has been confirmed in experimental and clinical studies on primary arterial hypertension as a result from changes of intracellular and extracellular zinc content. In arterial hypertension, the levels of zinc in serum, lymphocyte, and bone decrease while increasing in heart, erythrocytes, kidney, liver, suprarenal glands and spleen. These changes result in the loss of zinc homeostasis that leads to various degrees of deficiency, not entirely compensated by nutritional factors or increased absorption in the gastrointestinal tract [36].

Serum copper was a significantly high in CAD positive than CAD negative ones (Table 3) this was in consistence with other reports [37] contrary report to this had also been documented [23].

It is well known that copper plays a vital role in oxidative stress in which copper in its free form is a potent cytotoxic element because of its redox chemistry. It readily participates in Fenton and Heiber Weiss reactions to generate reactive oxygen species, a high level of copper enhances the toxic effect of metal dependent free radicals [38]. Diabetic CAD patients also had higher serum copper, our study was in corroboration with other reported studies [31,39-41] while unlike other [27] in which they found low serum copper in DM than non DM control.

Copper acts as a pro oxidant and may participate in metal catalyzed formation of free radicals [42]. The increased production of free radicals is likely to be associated with development of type 2 DM [41]. The increase in copper levels in patients with type 2 DM might be attributed to hyperglycaemia, which stimulates glycation and causes release of copper ions from copper binding sites of proteins. The release of copper ions into blood further accelerates the oxidative stress [43].

The above finding is true also for hypertensive patients, our study found higher serum copper in hypertensive CAD patients (Table 6), this finding is consistence with [44] and inconsistence with other report in which there was no statistical difference in the serum level of copper in hypertensive patients than control [31].

In a study done in Brazil [45] which revealed an elevated serum level of ceruloplasmin in hypertensive patients this could explain the higher level of copper in our patients hence ceruloplasmin is a copper containing enzyme.

Many epidemiological studies have demonstrated the relationship

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>Present n(%)</th>
<th>Absent n(%)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM</td>
<td>51(54.2)</td>
<td>43(45.8)</td>
<td>94</td>
</tr>
<tr>
<td>HT</td>
<td>56(59.5)</td>
<td>38(40.5)</td>
<td>94</td>
</tr>
</tbody>
</table>

Table 2: Classification of coronary artery disease patients risk factors.
of serum iron status and cardiovascular disease. However, contradictory results have been found [46,47], our result reveals that there was no significant changes in the serum level of iron between patient with CAD and others without CAD (Table 3), this finding has been substantiated by other studies [48-50] and in contrary with others [51,52] which were concluded that serum iron is higher in atherosclerotic heart disease and associated with its severity. One explanation of our result is the fact that human body has evolved a delicately balanced network to monitor iron entry, transport it to sites of need, and serve as a distinctive storage and recycling system, in the absence of an excretory system, to remove excess iron [53] on the other hand they found that stored iron concentrations, as assessed by serum ferritin, is a strong and independent risk factor for premature CAD [54], also many studies conclude that high serum ferritin increase the risk of atherosclerosis in absence of other risk factors, it act as a catalyzer in the production of oxygen free radical and lipid peroxidation and so lead to the formation of oxidized LDL [55].

Similarly, there was no statistical difference in the level of serum iron among diabetic and hypertensive CAD patients (Tables 5 and 6) this results were agreed with this study [56] while disagreed with others [27,44] which revealed that there was low serum iron level in diabetic and hypertensive patients respectively.

**Acknowledgements**

Thanks to God, blessings and greetings are up on the prophet Mohammed and Ahlulbyat. Special thanks to the staff in the biochemistry department/ College of Medicine and to the staff in AL-Basra Cardiac Center at Al-Sader Teaching Hospital for their kind support and cooperation. And I should not forget to thank all who help me without exception.

**Table 3: Biochemical parameters in the study population.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Obstructive CAD</th>
<th>Non CAD</th>
<th>P value**</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. Zinc(µg/dl)</td>
<td>56.60 ± 11.68</td>
<td>103.23 ± 20.62</td>
<td>0.0001</td>
</tr>
<tr>
<td>S. Copper(µg/dl)</td>
<td>171.27 ± 28.87</td>
<td>121.33 ± 28.52</td>
<td>0.0001</td>
</tr>
<tr>
<td>S. Iron (µg/dl)</td>
<td>113.33 ± 24.15</td>
<td>118.73 ± 23.95</td>
<td>0.115</td>
</tr>
</tbody>
</table>

*values expressed as mean ± SD **P value <0.05 consider statistically significant.

**Table 4: Logistic regression analysis adjusting for all major cardiovascular risk factors.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>OR</th>
<th>95% CI</th>
<th><em>P</em> value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lower</td>
<td>Upper</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zinc</td>
<td>0.743</td>
<td>0.648</td>
<td>0.838</td>
</tr>
<tr>
<td>Copper</td>
<td>1.094</td>
<td>1.067</td>
<td>1.123</td>
</tr>
<tr>
<td>Age*</td>
<td>1.080</td>
<td>0.85</td>
<td>1.31</td>
</tr>
<tr>
<td>Sex: *female</td>
<td>1.12</td>
<td>0.74</td>
<td>1.5</td>
</tr>
<tr>
<td>Female</td>
<td>2.04</td>
<td>0.65</td>
<td>3.43</td>
</tr>
<tr>
<td>BMI*</td>
<td>1.61</td>
<td>0.65</td>
<td>2.57</td>
</tr>
<tr>
<td>DM #</td>
<td>3.50</td>
<td>0.98</td>
<td>6.02</td>
</tr>
<tr>
<td>HT #</td>
<td>3.04</td>
<td>0.92</td>
<td>5.16</td>
</tr>
<tr>
<td>Smoking #</td>
<td>4.03</td>
<td>0.87</td>
<td>7.19</td>
</tr>
</tbody>
</table>

*P value <0.05 consider statistically significant. *^ values expressed as mean ± SD. *value express as n (%).
**Trace elements** | **DM** | **Non DM** | **P value**
--- | --- | --- | ---
Copper | 183.74 ± 30.34 | 156.47 ± 18.37 | 0.010
Zinc | 51.16 ± 10.39 | 63.05 ± 9.75 | 0.002
Iron | 113.29 ± 26.07 | 113.38 ± 21.96 | 0.987

*P<0.05 consider statistically significant.**Values expressed as mean ± SD.

Table 5: The effect of diabetes on trace elements in CAD patients group.

**Trace elements** | **Hypertensive** | **Normotensive** | **P value**
--- | --- | --- | ---
Copper | 177.65 ± 30.28 | 161.86 ± 24.09 | 0.035
Zinc | 53.82 ± 11.29 | 60.70 ± 11.15 | 0.007
Iron | 112.10 ± 24.09 | 115.14 ± 24.45 | 0.891

*P<0.05 consider statistically significant.**Values expressed as mean ± SD.

Table 6: The effect of hypertension on trace elements in CAD patients group.

References


