Study the activity of catalase, copper, ceruloplasmin and other biochemical features in first trimester Iraqi pregnant women
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ABSTRACT
Minerals trace elements, such as zinc, copper, iron, and several antioxidants like vitamins and ceruloplasmin, catalase, have significant influence on the health of pregnant women. The aim of the current study is to evaluate the relationship of trace elements, antioxidants and several antioxidants enzymes level in first trimester Iraqi pregnant women. The present study comprises of 50 normal pregnant women were selected in first trimester in attending for antenatal checkup to the hospital, and 30 non-pregnant women as control group. Venous blood (5 mL) was collected from pregnant women. In normal pregnancy, the copper and ceruloplasmin, total iron binding capacity (TIBC), and vitamin C concentration were statistically significantly increased in pregnant women group when compared to control group. The present study infers that iron is the most important micronutrient and it should be supplemented as a daily requirement in pregnant women. TIBC can be used as a diagnostic measurement and follow-up health status of women in pregnancy.

Keywords
Pregnancy, Catalase, Copper, Ceruloplasmin, Vitamin C, Vitamin E, Vitamin A.

Introduction
Pregnancy is associated with increased demand of all the micronutrients like iron, copper, and ascorbic acid [1]. The insufficiency of these nutrients could affect pregnancy, delivery and outcome of pregnancy. Vitamins and minerals are together referred to as micronutrients and have important influence on the health of pregnant women and growing fetus[2]. The two trace elements namely zinc, and copper are essential during pregnancy and these elements should be supplemented as a daily requirement in pregnant women [3,4]. Pregnancy is a condition exhibiting increased susceptibility to oxidative stress, defined here as a disturbance in the pro oxidant-antioxidant balance in favor of the former, leading to potential damage[3,6]. Pregnancy is characterized by dynamic changes in multiple body systems resulting in increased basal oxygen consumption and in changes in energy substrate use by different organs including the fetoplacental unit. From early pregnancy the human placenta influences maternal homeostasis; it is rich in mitochondria and when fully developed consumes about 1% of the basal metabolic rate of the pregnant woman [6].Serum ceruloplasmin is a copper transporting globulin synthesized in liver and possesses ferrooxidase activity. It acts as an antioxidant in serum by oxidizing ferrous iron which could otherwise act as a catalyst in generating toxic free radicals[7]. Pregnancy linked with significant physiological changes, which increases the require for iron. Iron is mainly important trace element and it also an necessary component of hemoglobin synthesis. Iron is required for additional erythrocyte production during pregnancy[8]. Catalase is an enzyme found in nearly active organisms uncovered to oxygen. It catalyzes the decomposition of hydrogen peroxide to water and oxygen [9].

Vitamin E is incorporated into the lipid portion of cell membranes and carrier molecules and protects these structures from toxic compounds, heavy metals, drugs, radiation and free radicals. Vitamin E also protects cholesterol from oxidative damage. Vitamin E plays an important role in the regulation of platelet function and might contribute to the maintenance of placental microcirculation by inhibiting platelet aggregation.

Because of its antioxidant effects, a diet high in vitamin E appears to be protective against common health conditions such as heart disease, cancer and strokes[10].

The role of vitamins C and E in preventing free radical damage is well known and their nutritional adequacy is important in pregnancy. Vitamin C deficiency affects placental structure and ROS and facilitates placental infection, all of which result in increased risk of premature rupture of placental membranes and premature births [11,12].

The total iron-binding capacity of serum (TIBC) has been observed to rise during the course of pregnancy. This rise in TIBC, coupled with a lowered percentage saturation of transferrin and a fall in hemoglobin concentration, has given rise to the concept of the "physiological anemia of pregnancy[7]."

The present study was done to assess the comparison of trace elements and serum catalase and ceruloplasmin level in first trimester iraqi pregnant women.

Material and Methods
The present study comprises of 50 normal pregnant women were selected in first trimester in attending for antenatal checkup to the hospital, and 30 non-pregnant women as control ranging in age mean± SD (26.4±3.4) years. The subjects with obesity, diabetes mellitus under medication and untreated diabetes, severely anaemic (<7.0gm% of Hb) and those suffering from any other systemic disorder were excluded from the study. Analysis of variance followed by taking 5-7.5 mL of blood was drawn by venipuncture and collected in a heparinized tube (10 units/mL of blood). The subjects were divided into the
two groups: group 1- thirty non-pregnant women, group 2-fifty pregnant women 1st trimester. Random blood samples were taken and the following parameters were estimated in both cases and control. The following parameters were analyzed within 10-15 minutes of collecting blood sample. Serum TIBC and iron level were measured by spectrophotometric methods supplied by Biolabo kit, France. The ceruloplasmin levels in human serum by turbidimetric method supplied by Fortress Diagnostics Limited. Vitamin C levels in serum were estimated by the method of Tietz [13]. Vitamin E levels were determined according to a modified of Hashim and Schuttringer[14]. The concentration of vitamin A in serum was determined according to a modified method of Neeld and pearson [15]. Catalase was determined using flame atomic absorption spectrophotometer type GBC 933 plus at $\lambda$ 324.7nm. This atoms are capable of absorbing light at the specific wavelengths of its line spectrum[17].To calculate the unsaturated iron binding capacity (UIBC), the serum iron concentration was subtracted from the TIBC.

$$\text{UIBC} = \text{TIBC} - \text{Serum iron concentration}$$

Transferrin can be estimated indirectly from the TIBC value by the following equation [18]:

$$\text{Transferrin (µg/dL)} = 0.7 \times \text{TIBC (µg/dL)}$$

The percentage of saturation of transferrin with iron is determined by the following equation :

$$\% \text{ Saturation of transferrin with iron} = \frac{\text{Serum iron}}{\text{TIBC}} \times 100$$

The women gave their oral and written consent before entering the study. All investigations were conducted in accordance with guidelines in the Declaration of Helsinki.

All statistical analyses in studies were performed using SPSS version 17.0 for Windows (Statistical Package for Social Science, Inc., Chicago, IL, USA). Descriptive analysis was used to show the mean and standard deviation of variables. The significance of difference between mean values was estimated by Student T-Test. The probability $p< 0.05 = \text{significant}$, $p> 0.05 = \text{non-significant}$. Correlation analysis was used to test the linear relationship between parameters ANOVA test was used to show the differences between variables of differentiated groups.

**Results and Discussions**

In normal pregnancy, the copper and TIBC concentration were statistically significantly increased ($p<0.001$). Serum ceruloplasmin and serum vitamin C concentration, were observed significantly raised ($p<0.01$) in normal pregnant compared to non-pregnant woman. On comparison of the normal and non pregnancy women , the serum level of vitamin A and level of catalase were observed not statistically significant variations ($p>0.05$), and level of vitamin E, serum iron were statistically significantly decreased ($p<0.05$, $p<0.01$) , respectively; in normal pregnant compared with control group as shown in table 1. There were A significant positive correlation between TIBC and vitamin C, vitamin A and copper and A significant negative correlation between TIBC with iron as shown in figure 1, while there wasn’t A significant correlation between TIBC and ceruloplasmin .

Confident alterations from the normal in copper, iron and TIBC of the serum have been observed during pregnancy. The increase in TIBC during the first trimester of pregnancy occur at a time when require for iron on the part of the fetus is great. The present explanations reveal a close time correlation between the decrease in serum iron and the increase in TIBC. The plasma transferrin rises earlier during pregnancy in those with concomitant iron deficiency[19] the administration of supplemental iron in this series has probably contributed to the later rise in plasma transferrin .The function significant of the increase in carrier protein, transferrin .and therefore TIBC is uncertain. It may be a consequence of increased estrogens during pregnancy stimulating the rise in TIBC, not only of iron but also of hormones like thyroxin and corticosteroids [20].

Iron excess and iron-mediated oxidative stresses have been demonstrated in the intestinal mucosa, liver, spleen, bone marrow and placenta. However, it is vulnerable to oxidative damage secondary to the continuous presence of a relatively small excess of iron intake [21]. The production of hydroxyl and methoxyl radicals in both the luminal and mucosal contents of the gastrointestinal tract verify the role of iron in free radical damage[21].

![Figure 1: The correlation between TIBC and several biochemical parameter](image-url)
The release of oxygen from motherly hemoglobin is preferential by the lower partial pressure of oxygen in the placental cellular structure and fetal circulation, which has a greater affinity for oxygen, and by the release of fetal and placental metabolites, which lower blood pH causing a displacement of the Hb dissociation curve favoring oxygen delivery [22]. As it matures and its vascularization develops, it changes to an oxygen-rich environment and its rich mitochondrial mass favoritism the production of reactive oxygen species (ROS), which increases free iron liberated from iron sulfur clusters [23].

Ceruloplasmin level was found to be elevated during pregnancy than in non pregnant women, suggesting an oxidative surroundings and stress[24,25]. Ceruloplasmin is part of a family of acute phase proteins that usually plays a defensive role in response to an immune infuritating stimulus. It is a multifunctional copper containing protein that was first isolated in blood in 1948 [23]. One of its main roles is as an antioxidant, as it has substantial ferrooxidase activity and can sequester other free radicals [26,27]. Serum levels of ceruloplasmin have been found to increase during normal pregnancy in some kind [28,29] and it is thought that this increase during gestation protects against oxidative stress associated with pregnancy[24,25]. Furthermore, an unusual ceruloplasmin profile could also be a sign of abnormal embryonic development. No studies exploratory the specific temporal pattern of change in ceruloplasmin during pregnancy in relation to fetal loss have been carried out in other species. However, there is some confirmation that either lower or higher levels of ceruloplasmin compared to levels observed during normal pregnancy could be a signal of problems associated with gestation in humans [24,30].

The non-significant statistically changes in catalase and vitamin A during the first trimester of pregnant women as seen in present study are similar to these reported previously study conducted to assess the level of catalase and vitamin A in pregnancy [31].

The present study showed that TIBC can be used as a diagnostic measurement and follow-up health status of women in pregnancy, also the current study recommends that follow-up of pregnant women by measuring the level of copper, as well as vitamin A, vitamin C, and vitamin E. Further studies are required to confirm these factors in other populations with folic acid supplement.

**Conflict of Interest**

The authors declares that there is no conflict of interests regarding the publication of this article

**Reference**


<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Pregnant women [n=50]</th>
<th>Control group [n=30]</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age(year)</td>
<td>27.2±3.0</td>
<td>26.4±3.4</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Copper (µg/dL)</td>
<td>157±13.1</td>
<td>119±18.0</td>
<td>&lt;0.001</td>
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<tr>
<td>ceruloplasmin(mg/dL)</td>
<td>41.4±6.0</td>
<td>35.2±6.8</td>
<td>&lt;0.01</td>
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<tr>
<td>Vitamin A (µmol/L)</td>
<td>1.0±0.1</td>
<td>1.0±0.2</td>
<td>&gt;0.05</td>
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<tr>
<td>Vitamin E(µmol/L)</td>
<td>18±3.4</td>
<td>21±4.5</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Vitamin C (µmol/L)</td>
<td>108±7.4</td>
<td>51±0.1</td>
<td>&lt;0.01</td>
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<tr>
<td>Catalase (KU/L)</td>
<td>50±1.3</td>
<td>48±1.0</td>
<td>&gt;0.05</td>
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<tr>
<td>Iron(µg/dL)</td>
<td>106±12</td>
<td>129±7</td>
<td>&lt;0.01</td>
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<td>TIBC (µg/dL)</td>
<td>316±49</td>
<td>288±28</td>
<td>&lt;0.001</td>
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<tr>
<td>UIBC(µg/dL)</td>
<td>208±35</td>
<td>156±30</td>
<td>&lt;0.001</td>
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<td>Transferrin (µg/dL)</td>
<td>221±30</td>
<td>203±28</td>
<td>&lt;0.01</td>
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<td>% Saturation of transferring</td>
<td>0.34±0.1</td>
<td>0.50±0.1</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>vitamin C and copper, ceruloplasmin, vitamin A, vitamin C, vitamin E, catalase, iron, TIBC and transferrin in sera of pregnant women and control group</td>
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