Serum trace elements and heavy metals in polycystic ovary syndrome

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Abstract
Objective: To study the serum levels of essential trace elements such as copper (Cu), zinc (Zn), manganese (Mn), magnesium (Mg), heavy metals, cadmium (Cd), cobalt (Co) and lead (Pb) in PCOS patients. METHODS: A total of 65 patients (35 patients with PCOS and 30 healthy controls) were included in the study. Serum levels of copper, zinc, manganese, lead, magnesium, carbon and cadmium were analyzed using the atomic absorption spectroscopic method. RESULTS: Serum copper and zinc levels were significantly higher, but manganese and lead levels were lower in patients with PCOS compared with healthy participants ($p < 0.01$). There was no difference between PCOS patients and controls with regard to serum levels of Mg, Co and Cd. CONCLUSIONS: Serum levels of trace elements and heavy metals may change in PCOS patients. The results of this study should be investigated with further trials in order to gain new insights into PCOS.
Keywords: Zink, Polycystic ovary syndrome, Chromium, Elements.

1. Introduction

Polycystic ovary syndrome (PCOS) is one of the most common endocrinopathies affecting approximately 6.5–8% of the reproductive age women (1,2). It is characterized by menstrual irregularities, clinical and/or biochemical hyperandrogenism and hyperinsulinemia (3). No one exactly knows what causes PCOS. Recent studies have indicated increased oxidative stress in the patients with PCOS (4,5).
The importance of essential trace metals in health and disease is indisputable because of their vital role in specific concentration ranges and toxicity at relatively high levels. Essential trace elements have four major functions as being stabilizers, elements of structure, essential elements for hormonal function and cofactors in enzymes (6).
Zinc (Zn) is essential for the structure of many enzymes and plays a fundamental role in approximately 300 enzymatic reactions. It has a critical role especially in the function of metalloproteins, inducing members of oxidoreductase, hydrolase, ligase and lyase family and has coactivating functions with copper (Cu) in superoxide dismutase (SOD) or phospholipase C (7). Manganese (Mn) and Cu are essential micronutrients incorporated into many metalloenzymes and proteins involved in cell metabolism and regulatory pathways controlling oxidative stress (7). Cadmium (Cd) is a heavy metal and may have a role in the formation of reactive oxygen species (ROS) (8). Lead (Pb) may also lead to the production of ROS by depleting glutathione (GSH) and protein-bound sulfhydryl groups and enhancing lipid peroxidation (9). Excessive generation of ROS is showed in patients with PCOS(4,10,11).We thought that there might be a relationship between trace elements, heavy metals and PCOS, linked to the oxidative stress. However, we have not encountered any study evaluating neither serum essential trace elements except magnesium (Mg) nor heavy metals in women with PCOS in the English literature.
In the present study, we aimed to investigate the serum levels of essential trace elements (Cu, Zn, Mn and Mg) and heavy metals (Cd, Pb and cobalt [Co]) in patients with PCOS.

2. Materials and Methods

Subjects

The study was conducted on 35 consecutive patients with PCOS (study group), body mass index (BMI) and age-matched healthy women (control group) from the outpatient clinics of the Department of Obstetrics and Gynecology and Al-Khansa Teaching Hospital in Nineveh Governorate.
The diagnosis of PCOS was based on the Rotterdam criteria, having two of the following three features - anovulation and/or anovulation, clinical and/or biochemical signs of hyperandrogenism and polycystic ovaries on ultrasound examination.
Age- and BMI-matched healthy women had regular menstrual cycles and did not have the signs of hirsutism. None of the subjects had been taking any drugs like insulin sensitizers, oral contraceptives, antiandrogens, glucocorticoids and mineral supplementations for at least 6 months before participating in the study. For both groups, the exclusion criteria included Cushing's syndrome, androgen-secreting tumor, diabetes, congenital adrenal hyperplasia, thyroid dysfunction and hyperprolactinemia. Weight and height were measured, and BMI was calculated as weight divided by height squared (kg/m2) for each subject.

Laboratory analysis

10 cc of venous blood was taken from each woman during the early follicular phase (between days 3 and 5) of the spontaneous menstrual cycle. Samples

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were kept in a cold box at +4°C and immediately transported to the laboratory. Serum samples were obtained by centrifugation of blood samples at 2500 rpm for 15 min at 4 °C and stored at −80 °C until analysis. Serum levels of copper, zinc, manganese, magnesium, carbon, cadmium and lead were measured by an atomic absorption spectrophotometer in the Department of Biochemistry. College of Education for Basic Sciences, University of Mosul. Atomic absorption spectrometric method is the best choice for measuring the amount of trace elements and heavy metals in blood serum due to its high sensitivity and specificity.

3. Statistical Analysis

Results were expressed as mean and SD. Student's t test was performed to evaluate differences between the group means. Pearson's correlation coefficient was used to evaluate the relationship among the variables and $p < 0.05$ were regarded statistically significant. Statistical analysis was performed using the Statistical Package for the Social Sciences for Windows software.

4. Results

Demographic characteristics and the comparison of serum levels of essential trace elements, heavy metals and hormones between the PCOS patients and control groups are shown in Table 1. There were no statistically significant differences between the two groups with respect to the serum Mg, Co and Cd levels. While the serum levels of Cu and Zn were higher, Mn and Pb levels were lower in PCOS patients than the controls ($p < 0.01$). In PCOS patients, while the mean serum Cu level was negatively correlated with BMI ($r = -0.710$, $p < 0.01$), serum Pb level was inversely correlated with total testosterone level ($r = -0.543$, $p < 0.01$). This relationship was not detected in healthy subjects.

Table 1: Trace element levels in the serum of women with PCOS compared to controls

	Patients with Pcos (n=35)	Controls (n=30)	P
BMI (Kg/m2)	22.65±3.10	23.63±3.80	0.43
Cu (Mg/ml)	0.72±0.12	0.63±0.11	0.0019
Mn (Mg/dl)	0.09±0.01	0.18±0.09	0.0019
Zn (Mg/ml)	0.91±0.22	0.76±0.18	0.0019
Mg (Mg/ml)	16.83±17.60	11.90±1.26	0.253
Pb (Mg/dl)	0.43±0.36	0.81±0.23	0.0019
Co (Mg/dl)	0.04±0.02	0.02±0.01	0.246
Cd (Mg/dl)	0.03±0.02	0.03±0.01	0.758

5. Discussion

To the best of our knowledge, this is the first study investigating the serum levels of trace elements (except Mg) and heavy metals in PCOS, which is a multifactorial disease, and its exact cause remains a mystery.

Cu is one of the essential trace elements for human body. In this study, we found higher serum Cu level in PCOS patients (normal range, 0.7–1.5 µg/mL) than the controls ($p < 0.001$). In addition, Cu level was negatively correlated with BMI in patients with PCOS. In the literature, there is no study analyzing the serum Cu level and its relationship with BMI in the patients with PCOS. Cu is a cofactor of many enzymes involved in redox reactions, such as cytochrome c oxidase, ascorbate oxidase or SOD. Besides its enzymatic roles, Cu has also a role in biological systems for electron transport (12). It can induce oxidative stress by catalyzing the formation of ROS and decreasing GSH levels (13,14). Many studies supported that increased oxidative stress may have a role in the pathogenesis of PCOS (10). GSH depletion, resulting from the increased production of ROS, was shown in patients with PCOS (15). Kuscu and Var (11) and Zhang et al. (4) also demonstrated higher SOD levels in PCOS patients. Therefore, there may be a relationship between Cu and PCOS. Padilla et al. showed that deficiency of Cu increased adiposity (16). We also found an inverse relationship between serum Cu level and BMI ($r = -0.71$, $p < 0.01$). However, PCOS patients in our study were nonobese and there was no significant difference between them and the controls with respect to the BMI. In addition, total number of PCOS patients and the controls was small. Therefore, we suggest further trials to be carried out to investigate serum Cu levels and its relationship with BMI in larger groups consisted of obese and nonobese patients with PCOS. Zn is another essential trace element and acts as a catalytic, structural and regulatory ion (17). In addition, it plays a role as an antioxidant by contributing to the structure of the Cu-Zn SOD and maintaining the metallothionein tissue concentrations (7). In our study, although we found that the mean serum Zn level was higher in patients with PCOS compared with the controls ($p < 0.05$), Zn levels remained within the normal range (0.60–1.10 µg/mL). Since this is the first study measuring the level of Zn in PCOS patients, further studies are needed to show whether it has a role in PCOS or not.

Mn is also an essential element protecting the body against oxidative stress and is a cofactor for the metalloenzyme, which is Mn-containing SOD (MnSOD) (18,19). It neutralizes the highly reactive superoxide ions to less reactive hydrogen peroxide (H2O2), which is followed by its immediate conversion to H2O by catalase and other peroxidases in the mitochondrial matrix (20). In this study, the mean serum Mn levels of PCOS patients were half of the corresponding serum levels of the controls (normal range 0.09–0.29 µg/dL). Since oxidative stress is increased in PCOS, it is possible that serum Mn level is decreased as a result of consumption in the antioxidant defense system including MnSOD.

Low level of Pb exposure has a number of negative consequences for human health (21). It damages cellular components by elevating levels of oxidative

stress (22). Although the level of Pb remained within the normal range (0–20 µg/dL), it was lower and there was an inverse correlation between serum Pb and total testosterone levels in patients with PCOS ($r = -0.543$, $p < 0.01$). In accordance with our results, Telisman et al. also showed an increase in serum testosterone level in men with occupational exposure to Pb (23). However, the numbers of subjects in both the study and control groups were few and there is no other study evaluating the serum Pb level and its relationship with testosterone in PCOS patients. Further trials are needed evaluating serum Pb levels in PCOS patients.

Mg is another essential element for human body. We found that the mean serum Mg levels in patients with PCOS and the controls were similar (normal range, 12–25 µg/mL; $p > 0.05$) and no correlation was detected between its levels and the age, BMI, testosterone, DHEAS and the other measured trace elements and heavy metals in our study. Although Muneyyirci-Delale et al. showed a lower serum Mg level in PCOS patients, Stefanidou M, Maravelias C, Dona A, Spiliopoulou C. Zinc: a multipurpose trace element. Arch Toxicol 2006; 80: 1–9. Stefanidou M, Maravelias C, Dona A, Spiliopoulou C. Zinc: a multipurpose trace element. Arch Toxicol 2006; 80: 1–9. Kauffman et al. noted no significant difference between PCOS patients and the controls with respect to its level, (24) which is in accordance with the results of our study. Similar to our findings, the results of both of these studies did not show any correlation between the serum level of Mg, steroid hormones, age and BMI.

Both Co and Cd may interfere with DNA repair process. Co may also induce DNA damage and mediate free radical generation (25). Cd is a heavy metal which is unable to generate free radicals directly but may contribute to the formation of ROS indirectly. It has also a role in the inhibition of gene expression and signal transduction (8). We found that there was no significant difference between serum Co (normal range, 0.1–1.2 µg/L) and Cd (normal range, <0.1 µg/dL) levels in PCOS patients and the controls. The levels of both elements were detected in normal range in each group. Therefore, we could not find any association between these elements and PCOS.

In conclusion, the serum levels of trace elements may change in patients with PCOS. The findings in this study should be investigated with further trials in order to obtain new insights into PCOS.

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