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Serum Copper/Zinc Concentration and Copper:Zinc Ratio in Some Selected Surgical Diseases

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ABSTRACT

The trace elements copper and zinc are crucial in the process of inflammation, tissue repair and wound healing. In this study, serum Cu and Zn concentrations were measured using Flame Atomic Absorption Spectroscopy in 25 adult patients with the following surgical conditions: Benign Prostate Hyperplasia (BPH) (8), Acute Appendicitis (AAP) (5), Hernia (4), Hydrocele (4), and Typhoid Perforation (TP) (4); and a control group of 10 age-matched, healthy individuals. Mean serum Cu and Zn concentrations were 109.0 ± 40.0 µg/dl and 100.0 ± 20 µg/dl, respectively, and the Cu/Zn ratio was >1 in the control group. All the surgical conditions were found to have a significantly lower serum Cu and Zn concentrations at $P < 0.01$. Using the non-parametric method of Friedman's One-Way Analysis of Variance (ANOVA), the highest decrease in serum Cu concentration (89.90%) was found in patients with hydrocele. Patients with typhoid perforation had the highest decrease in serum Zn concentration (82.0%). The Cu/Zn ratio was <1 in all the surgical conditions, with the exception of typhoid perforation. The Cu/Zn ratio of <1 found in the surgical diseases reported in this study can be used as a diagnostic aid in differentiating them from malignant/lymphoproliferative diseases where the Cu/Zn ratio is >2. Furthermore, these findings are suggestive of the need for the inclusion of Cu and Zn supplementation during the post-surgical management of benign prostate hyperplasia, acute appendicitis, hernia, hydrocele and typhoid perforation.

Keywords: disease, health, serum, surgery

INTRODUCTION

Trace elements have been reported to play important roles in health and disease (Underwood 1977; Prasad 1978; Shaw 1979; Harris 1997; Uauy *et al*. 1998). Copper (Cu) is an essential trace element for both humans and animals (Harris 1997). Cu performs many biological roles prominent among which are fuelling energy production, prevention of anaemia and bone disease, cellular anti-oxidant defense and prevention of inflammation (Shaw 1979; Conforti *et al*. 1983; Conlan *et al*. 1990; Linder and Hazegh-Azam 1996; Eaton-Evans *et al*. 1996; Turlund 1999). Studies have shown that serum Cu levels in malignant diseases increase in relation to disease activity (Cohen *et al*. 1984) with remission usually associated with return of Cu levels to normal ranges. Zn (Zn) is a constituent of more than 200 enzymes (Sgarbieri *et al*. 1999). It plays an important role in nucleic acid metabolism, tissue repair and growth, prevention of cells from oxidative damage and stabilization of protein structure (Halstead and Smith 1970; Cunane 1988; O'Dell 2000). Zn-finger motifs have also been found to regulate gene expression by acting as transcription factors (Rhodes and Klug 1993). This metal also plays an important role in cell signaling, hormone release, test perception, apoptosis, sperm production and immune response (Hambrigde *et al*. 1972; Prasad 1983; Troun-Tran *et al*. 2000). Blood Cu levels have been found to increase, and Zn levels decrease in patients with malignant lymphomas, lung cancer (Gupta *et al*. 1991; Oyama *et al*. 1994) breast cancer, chronic lymphocytic leukemia (Beguin *et al*. 1987; Gupta *et al* 1991) and Human Immunodeficiency Virus (HIV) infection (Libanor *et al*. 1987). In this study, we assessed the serum concentration of Cu and Zn and the Cu/Zn ratio in patients with the following surgical conditions: Benign prostate hyperplasia, acute appendicitis, hernia, hydrocele, and typhoid perforation, considering the important role these metals play in inflammation and wound healing.

SUBJECTS AND METHODS

Study design

The subjects involved in this study were selected by simple random sampling of surgical patients presenting at the Bauchi State Specialist Hospital. The patients consisted of individuals with Benign Prostate Hyperplasia (BPH), Acute Appendicitis (APP), Hernia, Hydrocele, and Typhoid Perforation (TP). Furthermore, the randomly selected subjects were screened to eliminate individuals with the following conditions: Burns, inflammatory bowel disease, chronic blood loss, pancreatic insufficiency, diarrhea, celiac disease, hepatic disease, diabetes mellitus, breast and lung cancer, alcoholism and cystic fibrosis because these are known to be associated with changes in serum Zn and Cu levels. Based on these selection criteria, twenty-five patients comprising of the following: (BPH) (8), (AAP) (5), Hernia (4), Hydrocele (4), and (TP) (4); (age range = 18–45) were found to be eligible for participation in the study. A control group of 10 age and sex-matched healthy adults were also enrolled in the study for comparative purposes.

Collection and preparation of serum samples

Blood sample (5 ml) was collected from each patient on admission using a sterile needle and syringe. Serum was prepared by transferring the blood sample into a clean, sterile, metal-free centrifuge tube. The sample was centrifuged at 3000 x *g* for 5 min and the supernatant serum sample removed by aspiration using a Pasteur pipette. The sample was then transferred into a clean, sterile, metal-free container and analyzed within 24 h. The control serum was prepared by collecting blood samples from 10 healthy male donors using the procedure described above.

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Short Communication

Assay for serum Cu and Zn concentration

The concentration of Cu and Zn in serum was determined by Flame Atomic Absorption Spectrophotometry (AAS) using Buck Scientific Atomic Absorption Spectrophotometer, VPG System, Model 210 (Buck Scientific Corporation, California, U.S.A.). Serum samples were digested using a mixture of nitric and perchloric acids. 0.5 ml of serum was mixed with 5 ml nitric acid and 2 ml perchloric acid. The mixture was heated for 2 hours at 100°C. After cooling, the digest was made up to 25 ml with doubly distilled, deionised water and used for the Cu and Zn determination.

Nitric acid (0.1 M) was aspirated for 30 seconds and then the absorbance of the instrument zeroed. After that, the absorbance for both standard and serum sample solutions were measured in triplicate readings, with aspiration of 0.1 M nitric acid for 10 seconds after each reading to clean the burner.

Statistical analysis

Data analyses were effected using MINITAB-10 Statistical Software. Comparison of mean serum Cu and Zn concentrations between the control group and patients were done using Friedman's non-parametric one-way analysis of variance (ANOVA). P values < 0.05 were considered significant.

Ethics

This work was conducted in accordance with the following ethical declarations: World Medical Association's Declaration of Helsinki (1996), World Medical Association's Declaration of Lisbon on the Rights of the Patient (1995), CIOMS/WHO International Guidelines for the Conduct of Research Involving Human Subjects (1993).

RESULTS AND DISCUSSION

The results obtained are shown in **Table 1**. The serum Cu concentration was found to decrease significantly in all the surgical patients, $P < 0.01$. The decrease ranged from 71.55% in patients with Benign Prostate Hyperplasia (BPH) to the highest drop of 89.91% in patients with hydrocele relative to the control serum Cu concentration of 109.0 ± 40.0 µg/dL. Serum Zn concentration was found to decrease within the range of 58.0% in patients with acute appendicitis (APP) to 82.0% in patients with typhoid perforation relative to the control serum Zn concentration of 100.0 ± 20.0 µg/dL, $P < 0.01$. The mean Cu/Zn ratio in the controls was 1.09. The Cu/Zn ratio in all the surgical patients was found to be <1 with the exception of typhoid perforation where the ratio was 1.11.

Generally, all the surgical conditions were found to lead to decreased serum Cu and Zn concentrations. Since these conditions involve varying degrees of tissue inflammation, this finding is not unexpected because inflammatory pathogenesis is usually associated with a decrease in the serum levels of some trace metals, particularly Zn (Davies 1984). All the conditions were found to present a serum Cu/Zn ratio of <1, relative to the control Cu/Zn ratio >1, with the exception of typhoid perforation. The plasma/serum Cu/Zn ratio has been reported to be a highly promising diagnostic aid as it reflects the level of metallothionein uptake of Zn (Webb and Cain 1982). This protein is responsible for maintaining Cu and Zn homeostasis. During infection or inflammation, macrophages release leukocyte endogenous mediator, which stimulates hepatic Zn uptake by serum metallothionein and caeruloplasmin (Powanda and Beisel 1982). The decreased serum Cu and Zn concentration in these surgical conditions may reflect an increased utilization of these metals in response to the underlying inflammation consequent to tissue damage and the need for their utilization in tissue repair, since they are both crucial to the wound-healing process (Turlund 1988; Sgarbeiri *et al.* 1999). Other studies have reported clinical conditions associated with a Cu/Zn ratio >2 (Delves *et al.*1973; Hrgovic *et al.* 1973; Beguin *et al.* 1987; Gupta *et al.* 1991, 1994). However to date, all the conditions reported are malignant or lymphoproliferative diseases associated with an excessive rate of cellular proliferative activity. Since the surgical conditions reported in this study are not malignant conditions, the lower serum Cu/Zn ratio (<1) can be ascribed to the differences in their pathogenesis relative to the malignancies. Although the serum Cu/Zn ratio in typhoid perforation was the only one found to be >1, the serum levels of both the metals are significantly lower than the control concentration. An impaired absorption capacity can be an additional factor responsible for the decrease, since it is a dysfunction of the gastrointestinal tract. The Cu/Zn ratio of <1 in these surgical diseases can be applied as a basis for differentiating disease conditions associated with malignancies and those whose pathologies do not involve tissue proliferation since it is the former that are found to have a serum Cu/Zn ratio >2. Furthermore, due to the significant decrease in both serum Cu and Zn concentration in BPH, AAP hernia, hydrocele, and TP, we recommend the use of Cu and Zn supplementation during the post-surgical management of these cases considering their critical role in tissue repair and the wound healing process.

Table 1 Mean serum copper, zinc concentration and Cu/Zn ratio in surgical patients and control (µg/dL).

Conditions	Mean serum Copper	Mean serum Zinc	Cu/Zn Ratio
Control	109.0 ± 40.0 *	100.0 ± 20.0 *	1.09
BPH	31.0 ± 14.0 *b	33.7 ± 11.0 *b	0.92
AAP	30.0 ± 20.0 *b	42.0 ± 17.0 *b	0.71
Hernia	20.0 ± 5.0 *b	29.8 ± 4.0 *b	0.67
Hydrocele	11.0 ± 1.0 *b	24.0 ± 3.0 *b	0.46
TP	20.0 ± 7.0 *b	18.0 ± 3.0 *b	1.11

* Values differ significantly at p < 0.01
b Value not significant at p ≤ 0.01

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