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Original Article

## Alterations in manganese level in the biological samples of young obese Saudi women



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### المخلص

**أهداف البحث:** هدفت هذه الدراسة إلى تقييم التغيير في مستويات المنجنيز في عينات أظافر الشابات البيديات.

**طرق البحث:** تم إجراء دراسة مستعرضة لعدد ٧٠ طالبة بين عامي ٢٠١٨ و٢٠١٩ في جامعة الإمام عبد الرحمن بن فيصل، المملكة العربية السعودية. حيث تم تحليل الأظافر لتقييم مستوى المنجنيز. كما تم تجميع بيانات شملت الخصائص الديموغرافية (العنوان والعمر والتعليم والحالة الاجتماعية)، إضافة إلى بيانات خاصة بنمط الحياة والعادات الغذائية مثل الاستهلاك المعتاد للمياه والحليب والمشروبات الغازية.

**النتائج:** شملت الدراسة ٣٠ يعانين من السمنة المفرطة و٤٠ يتمتعن بوزن طبيعي. كما قد بينت النتائج بأن مستوى المنجنيز لدى الفتيات البيديات كان أقل تركيزاً مقارنة بالمجموعة الأخرى بفارق إحصائي، وعندما تم دراسة التنبؤ بمستوى المنجنيز، وجد أن ممارسة الرياضة بانتظام واستهلاك الفاكهة والخضروات والأسماك واللحوم والماء قد يكون من العوامل المؤثرات المهمة في تحديد مستوى المنجنيز.

**الاستنتاجات:** أظهرت الدراسة اختلافاً كبيراً في مستوى المنجنيز في أظافر المشاركات اللاتي يعانين من السمنة المفرطة مقارنة بالمشاركات غير البيديات. هناك حاجة إلى مزيد من الدراسات لتحديد ما إذا كانت المرأة السعودية معرضة لخطر نقص المنجنيز.

**الكلمات المفتاحية:** نساء؛ بدانة؛ أظافر؛ المنجنيز؛ المملكة العربية السعودية

### Abstract

**Objectives:** This study aims to evaluate the changes in serum manganese levels in the nails of women with obesity.

**Methods:** A cross-sectional study was conducted between 2018 and 2019 at Imam Abdulrahman Bin Faisal University, KSA. It was conducted in a convenience sample of 30 women with obesity and 40 without obesity. We obtained biological samples of nails from the participants and analysed these samples using a plasma atomic emission spectrometer to estimate the levels of manganese. A standard questionnaire containing items related to demographic features, such as address, age, education, and marital status, was used. In addition, the data on the usual consumption of water, milk, and soft drinks during a day or week, eating habits, and other health information were included in the questionnaire.

**Results:** The results of this study show that manganese levels are significantly lower ( $p < 0.001$ ) in the group with obesity at  $0.34 \pm 0.06$  mg/kg than in the group without obesity at  $0.62 \pm 0.02$  mg/kg. Regular sports activity in a week and consumption of fruit, vegetables, fish, meat, and water are significant predictors of the levels of manganese in the body.

**Conclusion:** The study demonstrates a significant difference in the levels of manganese in the nails of obese participants compared to non-obese participants. Further studies are needed to determine whether Saudi women are at risk for manganese deficiency.

**Keywords:** KSA; Manganese; Nail; Obesity; Women

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## Introduction

Obesity is an abnormal accumulation of body fat. It is one of the major chronic health problems that is increasing daily. It has negative effects on work productivity, quality of life, and healthcare costs in both developed and developing countries.<sup>1</sup>

Based on World Health Organisation (WHO) estimates for 2016, the prevalence of obesity among females aged  $\geq 15$  years old in KSA was 39.5%.<sup>2</sup>

Literature reports that obesity is linked to several disorders such as diabetes, metabolic syndrome, respiratory diseases, cardiovascular diseases, hyperlipidemia, and malignancies.<sup>3,4</sup>

There is an increasing recognition of the impact of lifestyle changes and economic development on the prevalence of obesity.<sup>5</sup> Changes in the levels of some elements in the scalp hair have been associated with obesity.<sup>41</sup>

Manganese is an essential element with many biological benefits in nutrition and metabolism.<sup>6</sup> It acts as a co-factor in the central nervous system for a variety of enzymes, including those for neurotransmitter synthesis.<sup>7</sup> It is needed in anti-oxidant enzymes as well.<sup>8</sup> Manganese also plays a major role in the regulation of the immune system, blood clotting mechanism, reproductive process, cellular energy, and connective tissue and bone health.<sup>9</sup> It is essential in the protection of the body against oxidative stress.<sup>10</sup>

Analysis of trace elements in blood is not preferred because it does not reflect the actual amount in the human body due to homeostatic mechanisms.<sup>11</sup> However, biological samples such as nails have the advantage of “time-integrated measure of exposure time and body intake” due to their slow growth rate. Hence, it may be a better option to detect the concentration of trace elements in the body.<sup>12</sup>

An experimental study on mice subjected to high-fat feeding showed that manganese supplementation provided protection for beta-cell function and improved glycemic control.<sup>13</sup>

There are few studies that report the level of manganese in women with obesity.<sup>14,15</sup> In addition, these studies have shown inconsistent results.<sup>16</sup> In a study, serum manganese levels were found to be negatively correlated with serum insulin levels in obese diabetic women.<sup>17</sup> Moreover, a cross-sectional study reported a positive relationship between manganese level and obesity.<sup>18</sup>

Interestingly, manganese supplementation has been found to be important in reducing obesity and regulating lipid metabolism in the blood. A clinical study reported that manganese intake may protect against metabolic syndrome in women.<sup>19</sup> However, manganese deficiency accelerates the proliferation of vascular cells and damages the endothelium, thus increasing the risk of hypertension. In

addition, a significant association was found between manganese level and diabetes and/or prediabetes states.<sup>20</sup>

There are limited data on the influence of manganese concentration present in the nail samples of Saudi women on obesity, lifestyle factors, and dietary habits. Therefore, determining the manganese concentration may significantly contribute to a better assessment of the nutritional and health status of women with obesity.

This study evaluates the change in manganese levels and reports its predictors in nail samples of young women with obesity.

## Materials and Methods

A cross-sectional study was conducted from July 2018 to March 2019 using convenience sampling. A total of 70 female participants aged 19–30 years old studying at the Imam Abdulrahman Bin Faisal University (IAU) in KSA participated in the study. Those who participated in the study had natural nails, were non-smokers, and indicated themselves as healthy, without any chronic disease or cancer. Exclusion criteria included current pregnancy and a history of taking a trace element supplement in the past three months.

Participants were divided into two groups: group with non-obese (NOG) participants (body mass index [BMI] = 18.5–25 kg/m<sup>2</sup>) and obese (OG) participants (BMI > 30 kg/m<sup>2</sup>). All participants provided written informed consent. The study protocol was reviewed and approved by the Internal Review Committee of the IAU (IRB-2018-01-028).

A standard questionnaire was obtained and translated into Arabic (the language spoken by the participants). Through a pilot study, the instrument was found to be reliable and valid. Data related to demographic features (address, age, education, and marital status) were recorded. In addition, items such as sports activity (hours/week) fruits + vegetables (servings day) soft drink (servings week) fast food (servings week) water (liter per day) milk (liter per day) fish (servings week) chicken (servings week) meat (servings week) were included in the questionnaire.

Students were given a choice of responses ranging from 0 to a maximum of 7 days/week. For food thresholds, we calculated the proportion of students with a “healthy” intake of milk, vegetables, and fruit for  $\geq 5$  days/week and those with “unhealthy” nutritional options as consumption of health food for <3 days/week.<sup>21</sup>

### *Reagents and samples preparation procedure*

Nail specimens were collected in a dedicated plastic sack. The participants were instructed to remove any cosmetic products before cutting their nails. All samples were carefully stored at 25 °C until they were processed and analysed within 3 weeks of collection.<sup>22</sup>

A total of 5 mg of nail sample was washed in series with hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>, 30%), acetone, nitric acid (HNO<sub>3</sub>, 65%), and Triton-X 100 (Merck, Darmstadt, Germany). Sample dilution and digestion procedure was carried out by the method described by Ishak I and his colleagues in 2015.<sup>23</sup>

### Determination of manganese contents in samples

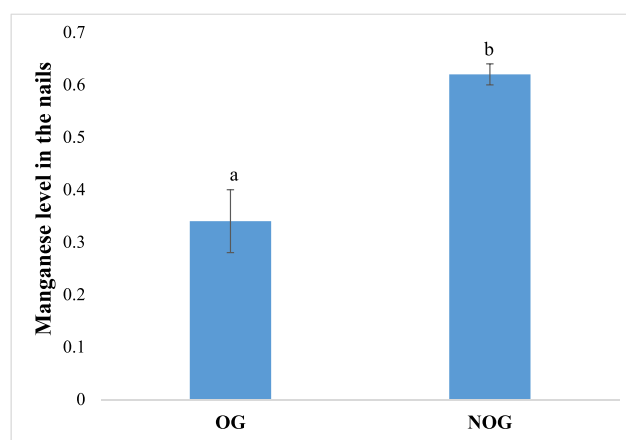
The samples used in this study were from human nails. Quantitative analyses of manganese were carried out using Inductively Coupled Plasma Emission Spectroscopy (ICP-OES-9800, ©Shimadzu Corporation, Japan), following the analytical methods and validation processes described by Sureda.<sup>22</sup>

### Statistical analysis

The data were analysed using SPSS for Windows version 25 (SPSS Inc., Chicago, IL, USA). The normality of the data distribution was assessed using the Shapiro–Wilk test. Data are presented as mean values  $\pm$  standard error of the mean (SEM). Percentages and proportions were used to describe categorical variables. Finally, a linear, logistic regression analysis was performed to identify the predictors of manganese levels in the OG. All tests were two-tailed, and a  $p$  value of  $\leq 0.05$  was considered statistically significant.

### Results

A total of 70 participants were enrolled in the current study: 30 in the OG and 40 in the NOG. The mean age of participants in OG and NOG was  $21.3 \pm 1.8$  and  $20.5 \pm 1.6$ , while the mean BMI was  $35.4 \pm 2.4$  and  $22.8 \pm 1.7$ , respectively. As expected, BMI was significantly different between



**Figure 1:** Content of manganese (mg/kg on a dry mass basis) in the nail samples of women with and without obesity. \* =  $p < 0.001$ ; NOG = non-obese group (n = 40; 57%); OG = obese group (n = 30; 43%).

the groups ( $p < 0.001$ ). Most of the participants were single (OG = 93.4% and NOG = 65%) (Table 1).

Figure 1 shows the manganese levels in the studied samples. Manganese level was significantly lower ( $p < 0.001$ ) in the OG ( $0.34 \pm 0.06$  mg/kg) than in the NOG ( $0.62 \pm 0.02$  mg/kg).

**Table 1: Basic and anthropometric characteristics of women with and without obesity.**

Characteristics	Groups		p value
	NOG	OG	
Age (mean + SEM)	20.5 $\pm$ 1.6	21.3 $\pm$ 1.8	0.23
BMI (mean + SEM)	22.8 $\pm$ 1.7	35.4 $\pm$ 2.4	<0.001
Income (n (%))			
$\leq 3000$ SR	40 (57.1%)	30 (42.9%)	0.19
3000–10000 SR	3 (100%)	0.0 (0.0%)	0.24
Marital status (n (%))			
Married	13 (32.5%)	2 (6.6%)	0.72
Single	26 (65.0%)	28 (93.4%)	0.51
Divorced	1 (2.5%)	0.0 (0.0%)	0.32

BMI = body mass index; NOG = non-obese group (n = 40; 57%); OG = obese group (n = 30; 43%); SR=Saudi riyals.

**Table 2: Lifestyle and dietary habits responses of women with and without obesity.**

Parameters	Groups		p value
	NOG	OG	
Sports activity (hours/week)	2.4 $\pm$ 0.3	1.9 $\pm$ 0.3	0.03
Fruits + Vegetables (servings/day)	4.2 $\pm$ 0.5	3.4 $\pm$ 0.5	0.72
Soft Drink (servings/week)	0.5 $\pm$ 0.1	0.7 $\pm$ 0.1	0.15
Fast Food (servings/week)	1.5 $\pm$ 0.2	2.0 $\pm$ 0.1	0.72
Water (litre/day)	1.4 $\pm$ .6	1.2 $\pm$ 0.6	0.82
Milk (litre/day)	1.7 $\pm$ 0.1	1.6 $\pm$ 0.1	0.04
Fish (servings/week)	1.7 $\pm$ 0.1	1.5 $\pm$ 0.1	0.51
Chicken (servings/week)	2.51 $\pm$ 0.1	2.5 $\pm$ 0.1	0.36
Meat (servings/week)	1.60 $\pm$ 0.1	1.5 $\pm$ 0.1	0.61

Values are shown as mean + standard error of the mean; NOG = non obese group (n = 40; 57%); OG = obese group (n = 30; 43%).

**Table 3: Predictors of monitored manganese in nail samples identified by multiple linear regression.**

Parameters	$\beta^*$	95% CI		<i>p</i> value
		lower	upper	
Sports activity (hours/week)	0.811	2.458	–5.25	0.02
Fruits + Vegetables (servings/day)	0.790	1.669	–7.12	0.01
Soft Drink (servings/week)	0.043	4.252	–3.538	0.73
Fast Food (servings/week)	–0.518	0.572	–3.069	0.10
Water (litre/day)	0.609	2.414	–7.615	0.01
Milk (litre/day)	0.058	4.028	–5.049	0.68
Fish (servings/week)	0.680	1.505	–8.444	0.02
Chicken (servings/week)	–0.158	3.592	–5.699	0.43
Meat (servings/week)	0.731	0.608	–10.068	0.04

$\beta^*$  = regression coefficient; 95% CI = 95% confidence intervals.

However, there were no significant differences in the dietary and lifestyle-related habits between females enrolled in NOG as compared to females in OG. A tendency toward healthier habits was detected among the NOG participants with regard to weekly activities and daily milk consumption ( $p < 0.034$  and  $p < 0.04$ , respectively) than among the OG participants, as shown in Table 2.

The following are significant predictors of manganese levels in the two study groups. Regular sports activity was measured through hours per week (Beta coefficient [ $\beta$ ] = 6.639,  $p < .022$ ), fruits consumed per day ( $\beta = 0.790$ ,  $p < .009$ ), water intake in litres (L) per day ( $\beta = 0.609$ ,  $p < .014$ ), fish servings per week ( $\beta = 0.680$ ,  $p < .025$ ), and meat servings per week ( $\beta = 0.731$ ,  $p < .040$ ). Soft drink, fast food, and chicken servings per week were not significant predictors ( $\beta = 0.043$ ,  $-0.518$ ,  $-0.158$ , non-significant). The overall model fit was  $F(13,2) = 27.370$ ,  $p < .036$ , with an  $R^2$  of .994 (Table 3).

## Discussion

The results reveal a marked decrease in manganese levels in the nails of young Saudi women with obesity. Our results are consistent with those of a previous research that demonstrated lowered serum level of manganese in females with obesity.<sup>15</sup>

The findings also show that NOG participants have adopted a healthy lifestyle and dietary habits compared to obese women.

The most important predictors for manganese levels in obese women are regular sports activity, consumption of water, and having considerable fruit, fish, and meat servings per week.

To date, few studies are conducted on the manganese levels in the nails of women in relation to their BMI.<sup>15,24</sup>

Manganese is a naturally occurring element that is available in low levels in food and water.<sup>25</sup> Interestingly, it is assumed that drinking water accounts for a small percentage of daily manganese consumption. Although this proportion may increase by as much as 20% depending on the geographic region and the source of water, the remaining manganese consumption is through food.<sup>25</sup>

Compared with OG, NOG participants significantly consumed more milk. This is consistent with other evidence

that Saudi women with obesity consumed lower amounts of milk.<sup>26</sup> However, milk does not seem to be a predictor of decreased manganese level in the study groups.

Our findings showed that consumption of soft drinks, fast food, and chicken are not among the predictors of manganese level alteration among the study groups. According to reported data, manganese intake is higher in vegetarians.<sup>27,28</sup> On the other hand, the same study has also indicated a positive correlation between manganese level and meat consumption.<sup>28</sup> The present study found that consumption of meat is a predictor for a higher manganese level in the OG than in NOG. However, our study design had a relatively small sample size; therefore, these results need to be confirmed in larger studies.<sup>29</sup>

The physical activity and nutrition of Saudi women are different from that of women in other countries.<sup>30,31</sup> There is an increased prevalence of obesity among Saudi female university students. Available data indicate that most college students did not consume fruits or vegetables frequently.<sup>32</sup> The majority (98.9%) skipped breakfast and reported having snacks.<sup>26,33</sup>

Available evidence indicates a decline in trace elements and malnutrition. “Low-cost, high-calorie, nutrient-poor foods” may play an essential role in obesity development.<sup>34</sup> However, these deficiencies may be responsible for the association between obesity and related metabolic diseases.

Modern food processing and agricultural techniques have led to a reduction in the levels of trace elements and vitamins in processed food. Despite the high-calorie intake, patients with obesity have relatively high rates of trace elements deficiencies.<sup>34</sup> The eating habits of this population segment in the Gulf region have changed from the consumption of grains, fresh fruits, and vegetables towards carbohydrate-rich diets.<sup>35</sup>

There is a limited number of reports from KSA that evaluate manganese intake in food. A recent study in KSA estimated the dietary intake of manganese in the main staple foods (chicken, meat, wheat, and rice).<sup>36</sup> The estimated manganese level was only 17% of the total recommended daily allowance set by the WHO.<sup>36</sup> Therefore, people in KSA may obtain the remaining amount of the total recommended daily allowance by consuming manganese-rich food, such as fish, nuts, tea, and grains.<sup>37</sup>



More importantly, the present data show that sports activity is significantly higher in the NOG and acts as a predictor of higher manganese levels. Data on the impact of sports activity on manganese levels is limited.<sup>38</sup> According to the WHO 2016 diabetes country profile, Saudi women were among those with the highest prevalence of physical inactivity in the world at 67.7%.<sup>2</sup> Scientific evidence has confirmed that physical activity is directly related to the trace element concentration in women's toenails.<sup>22</sup> However, it is well known that manganese is an integral compound of manganese superoxide dismutase enzyme that protects the body against the production of interleukin-1 beta, endogenous tumour necrosis factor alpha, and reactive oxygen species induced by sporting activities.<sup>39</sup>

The concentration of trace elements in nail samples reflects their actual concentration in the body for a longer period than that in body fluids.<sup>40</sup> This could serve as a good indicator for predicting any disturbance in homeostasis and aid in identifying links among factors that may lead to negative outcomes.

### Limitations

The findings of this study must be interpreted with caution because of its limitations. The dietary habits frequency questionnaire did not consider items related to the serving sizes of food that could affect the association between obesity and manganese levels. In addition, the study was cross-sectional; thus, we cannot infer causation from the current findings.

The possibility of a recall bias in the frequency of eating habits, sedentary behaviours, and physical activity cannot be entirely ruled out.

### Conclusion

Our study contributes to the small body of literature on the difference in manganese levels in women with obesity compared to women without obesity. Thus, metabolic derangement occurring in obesity may affect manganese status by redistributing it in the body, decreasing bioavailability, and increasing excretion.

### Recommendation

Further *in vivo* and *in vitro* studies are required to adequately estimate the association between manganese level alterations and metabolic parameters, as well as to highlight the mechanisms of manganese alteration and obesity in Saudi women. The management plan should include behavioural counselling to encourage women to be physically active and to reduce their consumption of food with high caloric content but with less nutritional value.

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### Conflict of interest

The authors have no conflicts of interest to declare.

### Ethical approval

The study protocol was reviewed and approved by the Internal Review Committee of Imam Abdulrahman Bin Faisal University (IRB-2018-01-028/31-1-2018).

### Authors' contribution

HMA and MTA conceived the idea, designed the study, conducted research, provided research materials, and collected and organised the data. HTA analysed and interpreted the data. HMA and MTA wrote the initial and final drafts of the article and provided logistical support. All authors critically reviewed and approved the final draft and are responsible for the content and similarity index of the manuscript.

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