Association between Iron Status and Lipid Peroxidation in Obese and Non-Obese Women

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Abstract

Background: Obesity is associated with increased lipid peroxidation. It has also been suggested that risk of lipid peroxidation increases with increasing body iron stores. The aim of this study was to examine the association of body iron status with the concentration of plasma malondialdehyde (P-MDA) as a marker of lipid peroxidation in obese and non-obese women.

Methods: In a case control study we investigated iron status by plasma ferritin, iron and total iron binding capacity (TIBC) measurements and lipid peroxidation by plasma malondialdehyde (MDA) levels measurements in 25 obese women and 25 non-obese women matched for age.

Results: Plasma ferritin levels were significantly higher in obese groups compared with control groups ($P< 0.001$). Plasma TIBC levels were not different in both groups and plasma iron levels were significantly higher in obese groups ($P< 0.05$). In obese groups, plasma MDA levels were significantly higher when compared with control groups ($P< 0.001$). There were positive correlation between body mass index and plasma MDA levels ($r= 0.75$, $P< 0.0001$). Plasma MDA levels were positively correlated with plasma iron levels ($r= 0.26$, $P= 0.001$) and plasma ferritin levels ($r= 0.39$, $P< 0.0001$) but not with TIBC levels.

Conclusion: These findings suggest that obese menstruating women are at low risk of depleting iron stores and hence, increasing body iron elevates the CHD risk by promoting the lipid peroxidation. Therefore, iron fortification programs might be undesirable for such subjects.

Keywords: *Iron status, Lipid peroxidation, Women, Obesity*

Introduction

Obesity is prevalent worldwide and is associated with increased mortality, increased cardiovascular diseased, diabetes and colon cancer. Although the exact biochemical mechanisms responsible for the association between obesity and the above diseases have not been completely elucidated, it is known that increase in the production free radicals associated with increased risk of above diseases (1). In the last decade evidence has accumulated that a crucial and causative role in the pathogenesis of atherosclerosis is played by the free radical process known as lipid peroxidation. It is currently believed that lipid peroxidation is involved in the oxidative modification of low density lipoprotein and this

ultimately results in the formation of atherosclerotic lesions (2, 3). Iron overload is usually associated with increased serum iron levels and decreased iron binding capacity (4). Generation of the reactive oxygen species (ROS) by iron-catalyzed Fenton reactions (5) have been implicated in the pathogenesis of many diseases including cancer, atherosclerosis and ischemia/reperfusion injury (6). It has been suggested that the risk of coronary heart disease increases with increasing body iron stores. In support of that hypothesis, a prospective epidemiologic study of heart disease in Finnish men found that the risk of heart attack increased with increasing levels of serum ferritin (7). The data also indicated that superoxide anions ($O_2^{\cdot -}$) can release iron from

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ferritin, providing free iron to catalyze the peroxidation of cell membranes (8). Although previous studies have shown that obesity is associated with increased iron stores in the body (9, 10), to the best of our knowledge there is no report in the literature of the effect of iron status per se lipid peroxidation in obese menstruating women. The purpose of this study was to test the hypothesis that iron status in obese women might be associated with increased lipid peroxidation. In this study we investigated iron status by plasma ferritin, iron and total iron binding capacity (TIBC) measurements in obese women and compared them to the matched control group. We also measured plasma malondialdehyde (P-MDA) concentrations product lipid peroxidation in obese group to study the association between iron status and P-MDA, a marker of oxidative stress.

Materials and Methods

The subjects used in this study were recruited from women under the cover of rural health centers of Kerman City, Iran. In this study, 160 menstruating women 20-45 yr old were randomly selected. Body weight was measured while the subjects were wearing light clothing without shoes to the nearest 0.1 kg. Height was measured to the nearest 1cm while subjects were not wearing shoes, in standing position. Body mass index (BMI) was calculated as weight (in kilogram) divided by height (in meters-squared) and was used to assess corpulence. From the data obtained, 25 subjects with BMI in the range of 30-40Kg/m^2 (cases) were chosen randomly and were age-matched with 25 subjects (controls) with healthy BMI (19-25Kg/m^2). In this study, pregnant and lactating women, blood donors, subjects with history of smoking, diabetes, hypertension, renal or liver diseases, and subjects who had taken iron or drugs that to modify their redox status were excluded.

Non-fasting venous blood samples were drawn from subjects between 8:00 and 12:00 a.m. Samples were collected into Vacutainer tubes containing ethylene diamine tetra acetic acid (EDTA).

Blood samples centrifuged at 3000 rpm for 10 min at 4º C and plasma was separated for the assay laboratory parameters. Subject's plasma was stored in -70º C until analysis. Plasma MDA concentrations were assayed by measurement of thiobarbituric acid reactive substances (TBARS) according to Satoh method (11). The pink chromogen produced by the reaction of thiobarbituric acid with MDA was measured at 530 nm. Plasma iron concentrations were assayed by atomic absorption method by use of autoanalyser. Plasma TIBC levels were measured by autoanalyser (Kodak Ektachem 500) using the method of colorimetric slide and standard kits. plasma ferritin concentrations were determined with chemiluminometric immunoassay method in the Ciba Corning ACS-180 analyzer.

We applied statistical power calculation and found that the number of participants studied was adequate to evaluate greater than 0.5 two-tailed standardized differences of the investigated concentrations of MDA between groups. In particular we achieved statistical power > 0.80 at< 0.05 probability level (*P* value).

For statistical analyses, data are expressed as mean±standard deviation (SD). Normality tests were applied using the Kolmogorov-Semirnov criterion. P-MDA and variables selected to estimate iron status were compared between obese and non-obese subjects by use of Student's *t*-test. The relationships P-MDA and variables selected to evaluate iron status, were analyzed through the calculation of the Pearson's correlation coefficients. Statistical significance was defined as a *P* value of less than 0.05.

Results

General characteristics of the study population (obese and non-obese groups) are shown in Table 1. Mean age was similar in obese and non-obese groups (Table 1). Plasma ferritin levels were significantly (*P*< 0.001) higher in obese group compared with the non-obese group (Table 2). Our data also revealed that obese women had higher plasma iron levels compared with

non-obese women ($P< 0.05$), as shown in Table 2. No significant difference was observed between study groups in plasma TIBC levels. Furthermore, [P-MDA levels were significantly ($P< 0.001$) higher in obese group compared with the non-obese group (Table 2).

Regarding P-MDA levels, we found that it was positively correlated with BMI ($r=0.75$, $P< 0.0001$), plasma iron levels ($r=0.26$, $P= 0.001$) and plasma ferritin levels ($r=0.39$, $P< 0.0001$) but not with plasma TIBC levels.

Table 1: General characteristics of study population (Data are presented as mean ± SD)

Variables	Non-obese (n=25)	Obese (n=25)	*P*-value
Age	32.4± 5.3	32.8± 6.7	0.72
Height (cm)	157.4± 5.9	158.6± 5.3	0.43
Weight (Kg)	53.3± 6	79.2± 7.2	<0.001
Body mass index (Kg/m^2)	21.5± 2.3	33.2± 2.8	<0.001

Table 2: Comparison of plasma iron, TIBC, ferritin and MDA levels in obese and non-obese groups

Variables	Non-obese (n=25)	Obese (n=25)	*P*-value
Plasma iron (μg/dl)	89 ± 41	110 ± 40	<0.05
Plasma TIBC (μg/dl)	319 ± 72	297 ± 73	0.28
Plasma ferritin (ng/ml)	32 ± 13	50 ± 25	<0.001
P-MDA (μmol/L)	1.4 ± 0.3	3.4 ± 0.7	<0.001

TIBC, Total iron binding capacity; P-MDA, Plasma malondialdehyde

Table 3: Correlations among BMI, iron status indicators and P-MDA (μmol/L) in all the subjects

Variables	Pearson's correlation coefficients (r)	*P*-value
BMI (Kg/m^2)	0.75	<0.0001
Plasma iron (μg/dl)	0.26	0.001
Plasma TIBC (μg/dl)	−0.09	0.36
Plasma ferritin (ng/ml)	0.39	<0.0001

BMI, Body mass index; TIBC, Total iron binding capacity

Discussion

In the present study, all means of the measures selected to estimated iron status were higher in obese than in non-obese women, except plasma TIBC levels. Obese women appear to have greater iron stores than do non-obese women, in terms of plasma ferritin and plasma iron concentrations. Our results were in agreement, however, with those of Fricker et al. (10), who found increased serum ferritin levels in obese women compared with non-obese women. In another study (12), greater BMI was associated with higher mean hemoglobin concentrations and hematocrits but not with higher mean serum iron concentrations. However, in this study serum ferritin was not measured. Because serum ferritin is the best indicator of iron stores (13) the higher concentration found in the obese group effectively shows that these subjects had higher iron stores than did the non-obese women. The

higher iron stores in the obese women might partly be related to dietary intake.

In the present study, we also measured lipid peroxidation product (MDA) in the subjects to investigate the association between measures selected to estimate iron status and MDA, marker of oxidative stress. Lipid peroxidation is a free radical-generating process which occurs on every membranous structure of the cell. Free radicals are known to be involved in a number of human pathologies including atherosclerosis (14), cancer (15) and hypertension (16). We observed that a plasma concentration of MDA was significantly higher in obese group than in non-obese group. In our study, plasma iron and ferritin levels were positively associated with P-MDA levels. Few studies have examined the association of plasma iron levels and P-MDA in human populations. Results from a study in patients with acute myocardial infarction showed an association of higher iron status with increased lipid peroxidation (17). In another study, results suggested that iron and copper status may be associated with lipid peroxidation in subjects without metal overload (18). Extensive reviews have been recently published concerning the role of iron in free radical reactions, such as lipid peroxidation (19, 20). Iron is largely stored in ferritin. Free metal ions can catalyze the formation of the highly reactive hydroxyl radical from superoxide and hydrogen peroxide (21). To promote free radical production, iron must be liberated from proteins, but body iron is so tightly bound that there may not be free iron available in vivo under physiological conditions. It is believed that oxidant stress itself can provide the iron necessary for formation of ROS, for example, by mobilizing iron from ferritin (19). No significant correlation was observed between plasma TIBC and P-MDA levels in our study. Yesilbursa et al. found, negative correlation between TIBC levels and MDA levels in coronary artery patients (13). In another study, Cooper and Liao found no relation between TIBC and incidence of coronary heart disease (22). Over 90% serum iron-binding capacity is accounted for by the iron transport protein transferring. Apart from its function as an iron-transporting protein, transferrin has long been known to be an antioxidant and its antioxidant property is believed to be related to its capacity to bind iron (19, 23). Menstruating women constitute a group at risk for iron deficiency (10) but, in this study we demonstrated that obese women be a group at low risk of iron deficiency. Modest long-term iron depletion was proposed as one of the factors protecting from coronary heart disease (CHD) (24) for its part, obesity was shown to be an independent risk factor for cardiovascular disease (25). The relationship between CHD and obesity might be partially linked to an increase in body iron stores.

These results suggest that obese menstruating women are at low risk of depleting iron stores, possibly because of high iron intake. Iron fortification programs might thus be undesirable in such subjects. Because iron deficiency and excess are both probably undesirable, it would be of great help to identify more precisely populations at risk of iron deficiency; iron supplementation could then be more personalized. Further studies are needed to clarify the relationships between obesity and iron status and to assess their consequences.

Acknowledgements

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