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Potential factors contributing to poor iron status with obesity

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Abstract Obesity rates continue to rise and iron deficiency continues to be the number one nutrient deficiency worldwide, and both can lead to significant adverse health issues. Furthermore, the factors contributing to the iron deficiency observed in obese subjects are not fully understood.

Aim of the work: Is to study the factors contributing to poor iron status in obese rats.

Materials and methods: Hemoglobin content, Hematocrite value (%), total iron binding capacity (TIBC), and transferrin saturation (TS%) were assessed in 20 obese and non obese female rats. Also Leptin, Interleukin-6 (IL-6) and systemic Hepcidin levels were measured in both groups. Serum ferritin levels were measured in additional 20 obese and non obese rats. The correlation between Hepcidin and different parameters and between ferritin and TS% was assessed.

Results: Serum Hepcidin levels, IL-6, serum ferritin and plasma Leptin were significantly high in obese group; also there was significant decrease in serum iron, TIBC and TS% ($P < 0.05$) in obese group compared with the non obese group. Difference in hemoglobin levels and Hematocrite values between both groups was not statistically significant. A direct correlation was observed between serum Hepcidin and body weight. Also a direct correlation between Hepcidin, and Leptin and IL-6 was observed.

Furthermore, there were significant inverse correlations between serum Hepcidin and serum iron and TS% and between ferritin and TS%.

Conclusion: IL-6 and Leptin may be part of the axis that links obesity, inflammation, and Hepcidin with poor iron status.

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1. Introduction

Poor iron status affects billions of people worldwide. The prevalence of obesity continues to rise in both developed and developing nations. An association between iron status and obesity has been described but, the mechanism explaining this relationship remains unknown.[1] Obesity is characterized by a state

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of low grade inflammation in different tissues.[2] Hepcidin is a small peptide hormone secreted by hepatocytes, circulating in blood plasma and excreted in urine.[3] Its production is increased in inflammation[4] and expression of Hepcidin in the liver is increased dramatically by inflammatory mediators[5,6] which may lead to iron deregulation. Another possibility is that Hepcidin may be linked to other adipokines commonly elevated in obesity including Leptin.[7] Furthermore, the factors contributing to the iron deficiency observed in obese subjects are not fully understood.

Thus the aim of the present work is to study the factors contributing to poor iron status in obese rats.

2. Materials and methods

This work carried out using 20 female Wistar rats weighing 200–225 g, purchased from the faculty of science Tanta University. During the study the animals were kept in wire mesh cages with access to water. The room temperature was about 22–24 °C and the animals were exposed to 12:12 h light dark cycles. Before start of work blood samples were taken from animals of both groups and assessed to exclude animals suffering from iron deficiency anemia. The animals were divided into two equal groups. Group 1 (non obese group) received only normal diet (rat chow) for 8 weeks.In group 2 (obese group), obesity was induced by feeding high fat diet which is composed of 70% fat, 20% carbohydrates and 10% protein. The meal consists of cooked caw fat, full cream milk, bread and green vegetables for 8 weeks.[8] The two diets of both groups are equal in amount but different only in there constituent with equivalent iron levels to avoid dietary iron deficiency. Normal and high fat diet constituents were purchased from El-Gomhoria Company, Cairo, Egypt. High fat diet was preserved at 4 °C until used. All protocols were approved by Tanta Faculty of medicine ethics committee.

A blood sample is taken from all these animals after 8 weeks for determination of Hemoglobin content by hemoglobin meter.[9] Hematocrite value was measured using a microcapillary reader (International Micro capillary Reader; International Equipment) following 3 min of centrifugation.[9] Serum iron and TIBC were measured according to method described by (Laycock et al.).[10] Serum ferritin was measured by using commercially available Enzyme Linked Immunosorbent assay kits (sigma).[11] TS% was calculated as iron/TIBC × 100. Interleukin-6 (IL-6) was immunoassayed using commercially available sandwich Enzyme Linked Immunosorbent Assay (ELISA) kit (ER3IL6, Pierce Endogen, Rockford, IL) for rats IL6, It detects up to below 16 pg/mL IL6 with intra-assay and inter-assay CVs ranging from 6.2–6.8% to 14.1–14.9%, respectively. Leptin was measured by commercially available ELISA kit.[12]

Serum Hepcidin levels in rats were measured by method described by (Murphy et al.).[13] We have measured serum ferritin, iron, TIBC and TS% in additional 20 obese and non obese rats.

Table 1 Comparison between control non obese group and obese group as regards different studied parameters.

Parameters	Control (non obese)	Obese
Body weight (gm)	213.6 ± 13.07	303.1 ± 18.27*
Hepcidin (ng/ml)	69.2 ± 6.78	266.6 ± 10.74*
Leptin (pg/ml)	223.7 ± 19.83	300.5 ± 5.97*
Serum iron (μg/dl)	188.2 ± 5.89	117.5 ± 6.98*
Ferritin (ng/ml)	61.9 ± 7.82	67.8 ± 9.45*
TIBC (μg/dl)	671.8 ± 7.07	592.2 ± 7.59*
TS%	27.9 ± 1.33	19.8 ± 1.77*
IL-6 (pg/ml)	126.2 ± 5.53	361.4 ± 7.68*
Hemoglobin (g/dl)	13.7 ± 0.41	13.4 ± 0.43
Heamatocrit value%	38 ± 1.63	37.6 ± 1.24

* Denotes statistical significance $P < 0.05$.

Table 2 Correlation between serum Hepcidin (ng/ml) and different studied parameters.

Hepcidin #(ng/ml)	*R*	*P*
Leptin	.941(**)	.000
TIBC (ug/dl)	−.986(**)	.000
IL-6 (pg/ml)	.997(**)	.000
TS%	−.925(**)	.000
Serum iron (ug/dl)	−.980(**)	.000
Body weight	.945(**)	.000

** Correlation is significant $P < 0.01$ level (2-tailed).

2.1. Statistical analysis

Data are expressed as means ± SD. Student's *t* test is used to compare between two groups.[14] Pearson's correlation coefficient test was used to correlate between parameters studied.[15] We considered statistically significant *P* value < 0.05.

3. Results

Table 1 revealed that obesity results in significantly lower serum iron, transferrin saturation (TS%), and total iron binding capacity (TIBC) ($P < 0.05$) if compared with non obese group, on the other hand there was significant increase in body weight, as well as the levels of serum Hepcidin, IL-6, ferritin and plasma Leptin ($P < 0.05$) in obese group when compared to non obese group. However the hemoglobin levels and Hematocrite values were statistically insignificant between obese and non obese group.

Table 2, Figs. 1 and 2 revealed that in obese group there was significant direct correlation between serum Hepcidin and body weight ($r = 0.945$; $P = 0.000$), statistically significant inverse correlations were found between serum Hepcidin and serum iron ($r = -0.980$; $P = 0.0001$) and TIBC (ug/dl) ($r = -0.986$; $P = 0.000$) and between serum Hepcidin and transferrin saturation ($r = -0.925$; $P = 0.0005$), also direct correlation between serum Hepcidin and IL-6 ($r = 0.997$; $P = 0.0001$) and a direct correlation between serum Hepcidin and plasma Leptin level ($r = 0.941$; $P = 0.0001$)were found.

Table 3, Fig. 3 revealed statistically significant inverse correlations between ferritin and TS%.

4. Discussion

Obesity is a major global health problem. Obesity and iron deficiency are two of the most common nutritional disorders worldwide. Several studies found higher rates of iron deficiency in obese than in normal-weight individuals.[16] In this study, we have observed that obesity is associated with poor iron status as evidenced by significant decreased serum iron, TIBC and transferrin saturation noted in obese compared to

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