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Elevated serum ferritin concentrations in prediabetic subjects

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Abstract

Background

Few data are available on the association of variables of the insulin resistance syndrome and serum ferritin, an indicator of body iron stores. We examined the relationship between serum ferritin levels and impaired fasting glucose, a pre-diabetes stage associated with insulin resistance, in this study.

Subjects & methods

One hundred and eighty seven people, including 91 subjects with impaired fasting glucose (IFG) and 96 healthy people who were well matched for age and sex, were enrolled. Body mass index (BMI) and blood pressure of the participants were measured and serum cholesterol, triglyceride, white blood cells (WBC) count, C-reactive protein (CRP) and ferritin were evaluated. All the data were analysed by *t*-test, χ^2 test and analysis of variance.

Results

The IFG group had higher serum ferritin concentrations (85.5±6.6 μg/L *vs.* 49.4±3.7 μg/L, p=0.001). A positive correlation was found between fasting plasma glucose and serum ferritin (r=0.29, p=0.001). Using multiple regression analysis, we found an association between serum ferritin and blood pressure (0.15, p=0.01), FPG (0.29, p=0.001), triglyceride (0.08, p=0.01) and cholesterol (0.07, p=0.03). The odds ratio for the association of IFG in male subjects with a high serum ferritin level was 8.3 (95% CI: 1.2–11.9, p=0.01) and for females was 3.06 (95% CI: 0.58–15, p=0.1).

Conclusion

Based on the data from our study, aelevation in serum ferritin can be seen in pre-diabetes stage, before the occurrence of an overt diabetes mellitus.

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Key words: ferritin, pre-diabetes, fasting plasma glucose, insulin resistance.

Introduction

Some epidemiological studies have reported a strong association between elevated serum ferritin concentrations and increased risk for diabetes.[1,2] Since publication of reports about the relationship between excess ferritin, coronary heart disease and insulin resistance, interest in ferritin as a possible risk factor for diabetes has increased. In our previous study in Iran, we revealed a positive correlation between type 2 diabetes mellitus and serum ferritin concentration.[3] If ferritin is considered to be a risk factor for type 2 diabetes, then it might be elevated in pre-diabetes stages, for example, in subjects who have impaired fasting glucose (IFG). This study was designed to investigate the association between serum ferritin concentration and IFG in Zanjan, a city about 300 km west of Tehran, in 2004.

Materials and methods

Subjects

The study was carried out in a group of subjects aged 20 years and above with IFG who had been identified in an epidemiological study in Zanjan, one of the provinces of Iran, in 2001. From a total of 2,200 people who had been randomly entered into the original study, 110 were recognised to have IFG, using two different fasting blood samples. They were recalled in 2004 to re-evaluate their fasting plasma glucose (FPG) concentrations, and all subjects with FPG more than 110 mg/dL (6.105 mmol/L) and less than 126 mg/dL (6.993 mmol/L) were enrolled. After excluding all subjects with haemoglobin concentration less than 12 mg/dL (7.4472 mmol/L) and those with acute or chronic inflammatory or infective disease, 91 subjects were entered into the study. The subjects were not on medications that might affect blood glucose concentration.

The control group consisted of 96 individuals who had normal blood glucose during the same epidemiological study in Zanjan in 2001 and whose FPG was normal (less than 100 mg/dL [5.55 mmol/L]) in the re-evaluation in 2004. These normal participants were matched with the cases for sex and age.

Approval was obtained from the ethics committee of Zanjan University of medical sciences. All the participants were informed about the goals of the study and informed consent was obtained from all of them.

Measurements

Weight was measured by the physician during the clinical

Table 1. Characteristics of the impaired fasting glucose (IFG) group and the normal plasma glucose group

Parameter	IFG group			Control group			*P value
	Male (n=41)	Female (n=50)	Total (n=91)	Male (n=43)	Female (n=53)	Total (n=96)	
Age (y)	45.7±14.5	49.4±16	47.7±16	45.7±14.5	49.4±16	47.7±16	0.9
Smokers (%)	24.4	12	17.5	25.6	3.8	13.5	0.5
Hypertension (%)	17	30	24	32.6	26.4	29	0.4
BMI (kg/m^2)	25.5±0.6	27±0.5	26.4±0.4	24.3±0.5	26±0.7	25.±0.5	0.8
FPG (mg/dL)	114.6±4.2	115±4.7	115±4.7	93±8.5	91.3±8.9	92±8.7	0.01
Cholesterol (mg/dL)	205±6.4	222±7.4	213±6.5	191±6.8	199±7.2	196±5	0.01
Triglyceride (mg/dL)	198±15	202±18	200±12	163±14	148±12	154± 9	0.002
Ferritin (μg/L)	108±11	67.7±7	85.5±6.6	59±5.5	41.5±4.8	49.4±3.7	0.0001
WBC	7,025±1,100	6,900±1,089	6,962±1,095	6,870±990	7,010±1,01	6,930±1,005	0.4
CRP (mg/L)	4.2±1.1	4.3±1.5	4.2±1.4	3.9±1.5	4.1±0.8	4±1.2	0.5

Key: FPG = fasting plasma glucose; BMI = body mass index; WBC = white blood cell (count); CRP = C-reactive protein
For continuous variables, data shown are mean ± SD
*p values calculated from the comparisons between the normal and IFG groups

Conversion factors: for cholesterol = 0.02586, for triglyceride = 0.01129 and for glucose = 0.0555

examination, using a balanced-beam scale and with the subject wearing light clothing. Height was measured by the physician using the clinic stadiometer and body mass index (BMI) was calculated based on weight divided by the height squared formula. Blood pressure was measured with all the subjects in a sitting position and with a standard manometer at two different visits, and the mean reading was taken as the blood pressure of the subject. Mean systolic blood pressure (SBP) > 140 mmHg or diastolic blood pressure (DBP) > 90 mmHg or current use of antihypertensive medications was defined as hypertension for the purposes of this study.

Laboratory measurements were performed at the laboratory of Zanjan University of Medical Sciences, Vali-e-asr Hospital, Zanjan, Iran. Plasma glucose was measured by the glucose-peroxidase colorimetric enzymatic method, with a sensitivity of 5 mg/dL and intra-assay coefficients of variation (CV) 1.7% for lower limit and 1.4% for upper limit concentrations. Inter-assay CV was 1.1% for lower limit and 0.6% for upper limit concentrations. Serum cholesterol and triglyceride of all the participants were measured after 14 hours of fasting, using a colorimetric method with a sensitivity of 5 mg/dL. Intra-assay and inter-assay CV for the assay were 1.6% and 1.1% for lower limit and 0.6% and 0.9% for upper limit concentrations, respectively. Complete blood count (CBC) was done for all the participants using haematological analyzer (Sysmex, KX-21, Toa.Co., Japan). C-reactive protein was measured by a high-sensitivity assay, using a latex particle-enhanced immunoturbidimetric assay with analytical sensitivity of 0.175 mg/dL; upper limit of normal was set at 5 mg/dL. Intra-assay and inter-assay CV for the assay were 0.6% and 0.9% for lower limit and 0.7% and 0.9% for upper limit concentrations, respectively. Serum ferritin was measured by ^{125}I coated tube immunoradiometric assay (Kavoshyar Co.kit). The assay was performed in duplicate. After adding 500 μL of ferritin tracer to 20 μL of the sample in anti-ferritin monoclonal antibody-coated tubes and incubation for one hour in room temperature with shaking at more than 280 rpm, radioactivity was measured in a gamma counter with the window adjusted for ^{125}I. The inter-assay coefficients of variation ranged from 2 to 4%. Ferritin concentrations more than 295 μg/L for men and 155 μg/L for women were defined as raised.[4]

Table 2. Correlations (r) between serum ferritin and other variables in all subjects in the study

Variable	Ferritin concentration (μg/L)	p value
BMI (kg/m^2)	0.06	0.4
BP (mmHg)	0.15	0.01
FPG (mg/dL)	0.29	0.001
TG (mg/dL)	0.08	0.01
Chol (mg/dL)	0.07	0.03

Key: FPG = fasting plasma glucose; BMI = body mass index; BP = blood pressure, TG = triglycerides; chol = cholesterol

Conversion factors: for cholesterol = 0.02586, for TG = 0.01129 and for glucose = 0.0555

Statistical methods

Results were analysed with SPSS version 11.5. Data are expressed as the means ± SD. Student's *t*-tests or χ^2 tests were used to compare clinical and laboratory data. Pearson regression and analysis of variance was used and the odds ratio was calculated for detection of a glucose-impaired condition in the presence of high serum ferritin concentration. Significance was considered at a level of p=0.05.

Results

In all, 187 people were studied, including 91 subjects with IFG and 96 persons with normal plasma glucose concentration. Table 1 shows the anthropometric and laboratory characteristics of the case and control subjects in this study. The cohorts were well matched for age and sex. There was no difference between the two groups for their white blood cell (WBC) count or C-reactive protein (CRP) level.

Serum ferritin was higher in the IFG cohort (85.5±6.6 μg/L *vs.* 49.4±3.7 μg/L, p=0.001). Levels of fasting plasma glucose in the IFG group were much higher than in the healthy control subjects. Table 1 compares various characteristics in the case and control cohorts. On comparing laboratory findings for male and female subjects, a significant difference was found only in their serum ferritin concentrations; in general, concentrations of ferritin in men were higher than in women (p<0.05). A positive correlation was found between FPG and serum ferritin in this study (r=0.29; p=0.001). Using multiple regression analysis, we found a significant correlation between serum ferritin and blood pressure, FPG, triglyceride and cholesterol but not with BMI (table 2).

The odds ratio for the association of IFG with high serum ferritin concentration was 3.3 (95% CI 1.3–8.3, p=0.01). The odds ratio for the association of IFG in male subjects with high serum ferritin level was 8.3 (95% CI 1.2–11.9, p=0.01) and for females was 3.06 (95% CI 0.58–15, p=0.1).

Subjects with hypertension had higher ferritin concentrations (71±4.9 μg/L *vs.* 55.6±5.2 μg/L in the normotensives, p=0.03). There was no significant difference between smokers and non-smokers in terms of their serum ferritin concentration (76.6±13 μg/L *vs.* 65.2±4 μg/L).

Discussion

In our study, the ferritin concentration in glucose-impaired subjects, a high-risk population for type 2 diabetes, was significantly higher than that in normal control subjects, implying that hyperferritinaemia occurs before elevation of plasma glucose concentration above 126 mg/dL (6.993 mmol/L).

In recent years, the association of high serum ferritin level and hyperglycaemia in type 2 diabetes has become a topic of interest.[4] Many studies have revealed elevated ferritin concentrations in diabetic patients in comparison with normal subjects.[3,5,6] One of these studies was reported from Zanjan, Iran.[3] Excessive ferritin concentration can be a marker of iron overload and subclinical haemochromatosis in diabetes patients. There are some reports of a link between C282Y and H63D mutations in the HFE gene (haemochromatosis gene) and type 2 diabetes.[7,8] Moreover, first-degree relatives of patients with type 2 diabetes mellitus with normal glucose tolerance have higher ferritin concentrations than normal control subjects.[5] These observations may suggest a genetic predisposition to hyperferritinaemia in type 2 diabetes.

Iron overload decreases insulin sensitivity[5,9] and can cause earlier complications in diabetes.[10] A small study proved that bloodletting, which resulted in a 50% reduction of serum ferritin concentrations, improved glycaemia and insulin sensitivity in patients with type 2 diabetes.[9] Therefore, in addition to pancreatic beta cell damage, insulin resistance may be the other explanation for hyperglycaemia following iron overload. Iron is a potent pro-oxidant, and reactive oxygen species have been shown to interfere with insulin signalling at the cellular level.[10] This effect, especially in the liver, may be the main mechanism of insulin resistance.[11] A recent review has shown that insulin resistance may be the cause rather than the consequence of disturbances in iron metabolism.[12]

Furthermore, abnormalities in ferritin metabolism following glycation in a hyperglycaemic state might be a primary cause of hyperferritinaemia in type 2 diabetes.[13,14] Glycosylated ferritin has a longer serum half-life[10] and glycaemic control itself influences serum ferritin concentrations.

Although we excluded all subjects with acute or chronic infection or inflammatory disorders and there was no significant difference between the two groups in our study in leukocyte count and CRP level, we cannot entirely exclude the possibility that the serum ferritin level is a marker of subclinical inflammation, which itself may be a risk factor for type 2 diabetes mellitus.[15] The reduced risk for diabetes in premenopausal women and vegetarian societies has been reported and may be explained by the iron overload hypothesis.[16,17]

Limited studies on patients with impaired glucose tolerance (IGT) have shown higher ferritin concentrations in this group and found a positive correlation between serum ferritin and 2-hour glucose concentration during glucose tolerance testing.[5] Both IFG and IGT are prediabetic states which are characterised by insulin resistance. These results suggest that hyperferritinaemia and iron overload may be the primary cause of insulin resistance before overt diabetes mellitus develops.

Conclusion

Reduced dietary iron intake, especially in men and postmenopausal women with additional risk factors for type 2 diabetes, may be advisable.[17]

Actively lowering body iron stores may be effective in preventing type 2 diabetes in selected subjects with impaired glucose metabolism.

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Conflict of interest statement

None declared.

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