

The relation between body iron store and ferritin, and coronary artery disease

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Original Article

Abstract

BACKGROUND: Iron is essential for many physiological processes; whereas, iron overload has been known as a risk factor in progression of atherosclerosis. The aim of this study was to investigate the importance of serum ferritin levels, which are known as an indicator of body iron stored in the incidence of coronary artery disease (CAD).

METHODS: In a case-control study, we evaluated 432 eligible men who underwent coronary angiography at Chamran Cardiology Hospital, Isfahan, Iran. They were separated into two groups of case (with CAD) and control (without CAD). All subjects had given written informed consents. Then, the blood samples were taken after 12-14 hours of fast by a biologist for measuring cardiovascular risk factors and body iron stores, including serum ferritin, serum iron, and total iron binding capacity (TIBC). For statistical analyses, chi-square test, Student's t-test, one-way ANOVA, and the logistic regression were used.

RESULTS: In the present study, 212 participants with CAD in the case group and 220 participants free of CAD in the control group were included in the analysis. At baseline, there were significant differences in serum ferritin ($P < 0.001$) and other cardiovascular risk factors between the two groups. Moreover, when other risk factors of CVD were included in the model, serum ferritin [Odd Ratio (OR) = 1.006, 95% confidence interval of 95% (95% CI) 1.00-1.01, $P = 0.045$] and serum ferritin ≥ 200 (OR = 4.49, 95% CI 1.72-11.70, $P < 0.001$) were associated with CAD.

CONCLUSION: High iron store, as assessed by serum ferritin, was associated with the increased risk of CAD. Furthermore, it was a strong and independent risk factor in the incident of atherosclerosis in the Iranian male population.

Keywords: Iron, Ferritin, Coronary Artery Disease, Coronary Angiography

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Introduction

Cardiovascular disease (CVD) is the single largest cause of mortality in the world and results from the combination of environmental and genetic factors.^{1,2} In this respect, though iron is essential for many physiological processes, iron overload has been known as a risk factor in progression of atherosclerosis.^{3,4}

Excessive iron is capable of stimulating the progression of atherosclerotic lesions, to catalyze the production of free radicals, and to promote lipid peroxidation by reducing the levels of antioxidants in plasma; therefore, it can be associated with the progression of atherosclerosis and increase in the

risk of ischemic cardiovascular events.^{5,6}

Epidemiological studies have provided contradictory results regarding iron stores and subsequent atherosclerosis and coronary artery disease (CAD).^{5,7,8} For example, Klipstein-Grobusch et al. observed an independent relationship between serum ferritin levels and carotid atherosclerosis.⁸ In addition, another study revealed that iron is an important factor in the process of atherosclerosis.⁹ However, Knuiman et al., with a 17-year follow-up study in Australia, evaluated the association between serum ferritin level and coronary heart disease (CHD) and stroke events. The results of their study

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did not show any evidence in relation to ferritin level as a risk factor for CVD.⁴ Another investigation proposed that excessive body iron stores are not associated with the risk of CHD in women.¹⁰

To our knowledge, in this respect no comparison was made between patients with CAD, according to the injured vessels, and individuals without CAD. We consequently undertook to further investigate the hypothesis of a link between iron and cardiovascular disease by analyzing the association of serum ferritin levels, as an indicator of body iron stores, with CAD in the Iranian male population, and comparing the differences between patients with CAD, according to the number of injured vessels, and individuals without CAD.

Materials and Methods

In a case-control study, we randomly evaluated 481 men who underwent coronary angiography at the Isfahan referral center for cardiac patients, Chamran Hospital, Iran, from May 2010 until January 2011. In this case-control study, participants were separated into two groups by simple sampling. The case group comprised of 223 patients with CAD if one or more coronary arteries had a stenosis $\geq 50\%$ and the number of significant stenosis vessels were also recorded. The control group comprised of 258 individuals without CAD according to their angiography data and if there was no significant epicardial artery stenosis. The risk of CAD was assessed by the cardiologist through watching selective coronary angiography. Of all of the participants, 432 were considered eligible for participating in the study, 212 in the case group and 220 in the control group. We excluded individuals with a recent history of surgery and acute or chronic inflammatory diseases, such as inflammatory bowel disease (IBD), and rheumatoid arthritis, gastric ulcer, cancer, viral or liver disease, and those who took iron and vitamin supplements. In addition, patients with thalassemia and hemochromatosis were excluded from the study. All subjects completed the questionnaire including medical history and smoking after giving a written informed consent. Mean diastolic and systolic blood pressures were calculated from two independent measurements, each of which was taken with the subject in a supine position after 10 minutes of rest. The blood samples were taken after 12-14 hours of fasting for measuring hematologic indexes, fasting blood sugar, and serum lipids by standard clinical laboratory procedure. Serum lipids, including triglycerides (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C) using enzymatic

methods, and low density lipoprotein cholesterol (LDL-C), were calculated according to the Friedewald et al. formula.¹¹ Serum samples were collected from the case and control subjects simultaneously and frozen at -20°C until used to determine serum ferritin levels, serum iron, and total iron-binding capacity concentrations (TIBC). Serum ferritin concentration was determined by enzyme-linked immunoassay (Ideal Company). The CVs were 2.8%, 4.0%, and 10.4% for ferritin concentrations of 389, 139, and 27 mg/l, respectively. Serum iron and TIBC were determined by photometry with an eppendorf patient oriented system (EPOS) Chemistry Analyzer. The research protocol was taken under the medical ethics standards and was approved by the Medical Ethics Committee of Isfahan University of Medical Sciences.

Chi-square test and Student's t-test were used to compare case and control groups. For considering the differences in the variables in the patients with the number of involved arteries, one-way ANOVA was employed. The logistic regression was used to estimate the incidence of CAD as dependent variable and serum ferritin as independent variable, adjusted for age, hypertension, diabetes, hyperlipidemia, and smoking. Numerical values were expressed as mean \pm standard deviation. P-values less than 0.05 were considered as statistically significant.

Results

In the present study 432 men were evaluated; they were divided into individuals with CAD ($n = 212$, mean age = 58.76 ± 11.01 years) and individuals without CAD ($n = 220$, mean age = 52.16 ± 12.72). Chi-square test and Student's t-test were used to compare case and control groups (Table 1). Significant differences were seen in ischemic heart disease, diabetes, hyperlipidemia, hypertension, smoking, and serum ferritin between the two groups. However, significant differences were not observed in other variables. In addition, we compared patients with CAD according to the number of injured arteries. In this section, we employed one-way ANOVA and outcomes showed that these three groups (injury in one vessel, two vessels, and three vessels) did not have any significant differences in serum ferritin, serum iron, and serum total iron binding capacity (TIBC) (Table 2).

Results revealed that CAD was associated with serum ferritin levels when age, diabetes, hypertension, hyperlipidemia, and smoking were included in the model. It showed that the case group that had serum ferritin ≥ 200 had a four-fold higher risk of atherosclerosis than the control group (Table 3).

Table 1. Basic characteristics of subjects in case and control groups

Variable	Case group n = 212	Control group n = 220	P
Hypertension (%)	70 (33.0%)	42 (19.0%)	< 0.001
Diabetes mellitus (%)	57 (26.7%)	25 (11.3%)	< 0.001
Hyperlipidemia (%)	78 (36.7%)	44 (20.0%)	< 0.001
Smoking (%)	145 (66.7%)	37 (16.7%)	< 0.001
Serum ferritin (mg/dl)	206.8 ± 156.3	147.3 ± 132.9	< 0.001
Serum iron (mg/dl)	106.8 ± 46.9	107.6 ± 29.6	> 0.050
Serum TIBC (mg/dl)	310.8 ± 99.6	337.7 ± 56.5	> 0.050
Transferrin saturation (%)	33% ± 22.0	29% ± 21.0	> 0.050

TIBC: Total iron binding capacity

Table 2. Comparison of the patients according to the injured vessels

Variable n = 212	One vessel n = 74	Two vessels n = 39	Three vessels n = 99	P
Serum ferritin (mg/dl)	236.5 ± 173.69	203.66 ± 199.34	187.78 ± 131.54	> 0.05
Serum iron (mg/dl)	118.9 ± 41.11	79.33 ± 46.23	110.07 ± 49.40	> 0.05
Serum TIBC (mg/dl)	341.2 ± 110.89	307.00 ± 130.00	290.78 ± 77.46	> 0.05

TIBC: Total iron binding capacity

Table 3. The association of ferritin and coronary artery disease

Variables	OR (95% CI)	P
Serum ferritin	1.006 (1.00-1.01)	0.045
Serum ferritin ≥ 200 ng/ml	4.49 (1.72-11.70)	0.001

Adjusted for age, hypertension, diabetes, hyperlipidemia, and smoking

OR: Odds ratio, CI: Confidence interval

Discussion

Ferritin is an iron storage protein. Serum ferritin concentrations are directly proportional to intracellular ferritin concentrations; therefore, it is considered to be the best indicator of body iron stores. This case-control study revealed that excess serum ferritin is associated with atherosclerosis in Iranian males with coronary artery disease. In addition, findings showed that the risk of high serum ferritin persisted when other risk factors such as age, hypertension, diabetes, hyperlipidemia, high LDL-C, and smoking were adjusted in the model. Moreover, results indicated that men with CAD and serum ferritin concentration ≥ 200 ng/ml had a four-fold higher risk of atherosclerosis than healthy men. Therefore, elevated serum ferritin level may have an independent adverse effect on the incidence of atherosclerosis in patients with CAD. No significant statistical difference was seen in patients with single vessel, double vessels, and triple vessels disease regarding serum iron, serum ferritin, and serum TIBC. Furthermore, lack of a significant statistical difference in the other parameters such as serum iron, TIBC, and transferrin saturation, which are involved in iron homeostasis, can be attributed to the laboratory errors, the diurnal changes, and the hemolysis of blood samples.¹⁰⁻¹²

Many epidemiological studies have considered the association of iron status and CVD; however, contradictory results have been presented. Haidari et al. concluded that high stored iron concentration, as assessed by serum ferritin, is a strong and independent risk factor for premature CAD in the male Iranian population.¹³ Other studies proposed elevated serum ferritin concentrations to be associated with increased risk of CVD and myocardial infarction in elderly population, and that it is the leading cause of death and illness in the world.^{8,14} A review article suggested strong epidemiological evidence is available that iron is an important factor in processing of atherosclerosis.⁹ Salonen et al. demonstrated that a ferritin concentration ≥ 200 mg/l was associated with a 2.2-fold increase in the risk of acute myocardial infarction in men after adjustment for other risk factors.¹⁵

Many studies suggested that elevated serum ferritin increased the risk of atherosclerosis in the presence of other risk factors. Ferritin can act as a catalyzer in the production of oxygen free radicals and lipid peroxidation and play a role in the formation of oxidized LDL.¹⁶⁻¹⁸

Oxidation of LDL causes the accumulation of lipids in endothelial and smooth cells, and prevents macrophages from leaving the arterial wall. Thus,

these effects promote the atherosclerosis lesion.^{1,2,9}

On the other hand, there are many investigations that were inconsistent with our findings. Armaganijan and Batlouni suggested that serum ferritin and other organic iron indicators were neither risk factors nor risk markers for coronary atherosclerosis and serum iron levels were higher in the group without atherosclerosis.¹⁹ Auer et al. showed that higher ferritin concentrations and transferrin saturation levels were not associated with an increased extent of coronary atherosclerosis in patients who referred for coronary angiography.⁷

It could be mentioned that these conflicting results may be due to the large variability in estimates of iron stores, which included serum iron, serum ferritin, serum transferrin, and etcetera, the diversity methods in the diagnosis of atherosclerosis, and the variability in the size of demographic samples.¹⁹⁻²¹

The strengths of this study are the large sample size and the exclusion of participants with known diseases and supplementations usage at baseline which reduced potential for some biases. The limitation of this study is that it was conducted in men only and thus the results may not be generalized to women.

In conclusion, high stored iron concentration, as assessed by serum ferritin, was associated with the increased risk of CAD, while the number of injured vessels in these patients did not have any association with the progression of disease. In addition, it should be noted that high stored iron concentration was a strong and independent risk factor in the incident of atherosclerosis in the Iranian male population.

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This article is derived from the thesis of an internal medicine resident.

Conflict of Interests

Authors have no conflict of interests.

References

- Meyers DG. The iron hypothesis: does iron play a role in atherosclerosis? *Transfusion* 2000; 40(8): 1023-9.
- Bozzini C, Girelli D, Tinazzi E, Olivieri O, Stranieri C, Bassi A, et al. Biochemical and genetic markers of iron status and the risk of coronary artery disease: an angiography-based study. *Clin Chem* 2002; 48(4): 622-8.
- Day SM, Duquaine D, Mundada LV, Menon RG, Khan BV, Rajagopalan S, et al. Chronic iron administration increases vascular oxidative stress and accelerates arterial thrombosis. *Circulation* 2003; 107(20): 2601-6.
- Knuiman MW, Divitini ML, Olynyk JK, Cullen DJ, Bartholomew HC. Serum ferritin and cardiovascular disease: a 17-year follow-up study in Busselton, Western Australia. *Am J Epidemiol* 2003; 158(2): 144-9.
- Kiechl S, Willeit J, Egger G, Poewe W, Oberhollenzer F. Body iron stores and the risk of carotid atherosclerosis: prospective results from the Bruneck study. *Circulation* 1997; 96(10): 3300-7.
- Zheng H, Cable R, Spencer B, Votto N, Katz SD. Iron stores and vascular function in voluntary blood donors. *Arterioscler Thromb Vasc Biol* 2005; 25(8): 1577-83.
- Auer J, Rammer M, Berent R, Weber T, Lassnig E, Eber B. Body iron stores and coronary atherosclerosis assessed by coronary angiography. *Nutr Metab Cardiovasc Dis* 2002; 12(5): 285-90.
- Klipstein-Grobusch K, Koster JF, Grobbee DE, Lindemans J, Boeing H, Hofman A, et al. Serum ferritin and risk of myocardial infarction in the elderly: the Rotterdam Study. *Am J Clin Nutr* 1999; 69(6): 1231-6.
- de Valk B, Marx JJ. Iron, atherosclerosis, and ischemic heart disease. *Arch Intern Med* 1999; 159(14): 1542-8.
- Sun Q, Ma J, Rifai N, Franco OH, Rexrode KM, Hu FB. Excessive body iron stores are not associated with risk of coronary heart disease in women. *J Nutr* 2008; 138(12): 2436-41.
- Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972; 18(6): 499-502.
- van der AD, Marx JJ, Grobbee DE, Kamphuis MH, Georgiou NA, van Kats-Renaud JH, et al. Non-transferrin-bound iron and risk of coronary heart disease in postmenopausal women. *Circulation* 2006; 113(16): 1942-9.
- Haidari M, Javadi E, Sanati A, Hajilooi M, Ghanbili J. Association of increased ferritin with premature coronary stenosis in men. *Clin Chem* 2001; 47(9): 1666-72.
- You SA, Wang Q. Ferritin in atherosclerosis. *Clin Chim Acta* 2005; 357(1): 1-16.
- Salonen JT, Nyyssonen K, Korpela H, Tuomilehto J, Seppanen R, Salonen R. High stored iron levels are associated with excess risk of myocardial infarction in eastern Finnish men. *Circulation* 1992; 86(3): 803-11.
- Balla J, Vercellotti GM, Jeney V, Yachie A, Varga

- Z, Jacob HS, et al. Heme, heme oxygenase, and ferritin: how the vascular endothelium survives (and dies) in an iron-rich environment. *Antioxid Redox Signal* 2007; 9(12): 2119-37.
17. Liao X, Lv C, Zhang X, Masuda T, Li M, Zhao G. A novel strategy of natural plant ferritin to protect DNA from oxidative damage during iron oxidation. *Free Radic Biol Med* 2012; 53(2): 375-82.
18. Kraml P, Potockova J, Koprivova H, Stipek S, Crkovska J, Zima T, et al. Ferritin, oxidative stress and coronary atherosclerosis. *Vnitr Lek* 2004; 50(3): 197-202.
19. Armaganijan D, Batlouni M. Serum ferritin levels and other indicators of organic iron as risk factors or markers in coronary artery disease. *Rev Port Cardiol* 2003; 22(2): 185-95.
20. Eftekhari MH, Mozaffari-Khosravi H, Shidfar F, Zamani A. Relation between Body Iron Status and Cardiovascular Risk Factors in Patients with Cardiovascular Disease. *Int J Prev Med* 2013; 4(8): 911-6.
21. Jankowska EA, Malyszko J, Ardehali H, Koc-Zorawska E, Banasiak W, von HS, et al. Iron status in patients with chronic heart failure. *Eur Heart J* 2013; 34(11): 827-34.

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