



# Serum ferritin is associated with carotid atherosclerotic plaques but not intima-media Thickness in patients with abnormal glucose metabolism



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## ABSTRACT

**Background:** We investigated the association between serum ferritin and carotid artery lesions in populations with abnormal glucose metabolism.

**Methods:** We included 70 participants with abnormal glucose metabolism and 170 participants with normal glucose metabolism and measured their baseline serum ferritin levels. During follow-up carotid intima-media thickness and carotid plaque were evaluated.

**Results:** Serum ferritin levels were higher in the participants with abnormal glucose metabolism ( $p < 0.01$ ). We further divided the patients with abnormal glucose metabolism into subgroups with and without intima-media proliferation, and found that ferritin was excluded from the final equation in the logistic regression. Furthermore, age, waist circumference, ferritin, 2h-PG, and total cholesterol were significantly different between the subgroups with and without carotid plaque. When the above data were included in a logistic regression model, the  $p$  values obtained for age, ferritin, and 2h-PG were 0.004, 0.032, and 0.011, respectively.

**Conclusions:** In the Chinese population, serum ferritin levels are significantly increased in patients with abnormal glucose metabolism. The carotid intima-media thickness showed no independent relationship with serum ferritin in patients with abnormal glucose metabolism. However, high serum ferritin is an important risk factor for carotid atherosclerosis in these patients.

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## 1. Introduction

Cardiovascular disease (CVD) is a major disease and is a leading cause of death worldwide. A total of 598 million of Americans died from heart disease in 2010, which corresponded to 24.2% of total deaths and was the primary cause of death in that year [1]. Heart disease has also begun to attract more attention in developing countries in recent years. In China, the rate of deaths from heart disease has increased in recent years rising to 21.6% in 2013. CVD is a common phenomenon that is responsible for 17% of all US medical expenditures [2]. Diabetes is an important risk factor for CVD and influences almost all of the pathologies associated with CVD, which range from the formation of atherosclerosis to cardiac death. Moreover, in the Chinese population, the prevalence of diabetes and prediabetes is high at 11.6% and 50.1%, respectively [3]. It is therefore imperative for researchers to identify a biomarker for CVD in diabetic patients.

Epidemiological studies have shown that excess iron is positively correlated with reactive oxygen species (ROS) [4], dyslipidemia, and insulin resistance [5,6], and that it may play a vital catalytic role in the

development of atherosclerosis [7]. Furthermore, subjects with abnormal glucose metabolism possess higher serum ferritin levels than those individuals with normal glucose metabolism [8,9]. However, little research has been conducted to examine the correlation between serum ferritin levels and atherosclerosis in subjects with abnormal glucose metabolism.

The aim of this study was to compare the baseline serum ferritin concentration in subjects with abnormal glucose metabolism and participants with normal glucose levels. Additionally, we investigated the association between serum ferritin and carotid artery lesions detected through carotid artery ultrasonography in populations with abnormal glucose metabolism during the follow-up period in 2012.

## 2. Materials and methods

### 2.1. Subjects and study protocol

Between 2007 and 2008, the Chinese Diabetes Society conducted a cross-sectional survey using a multistage, stratified sampling method to investigate the prevalence of type 2 diabetes and its complications in China. At that time, a total of 596 subjects from the Longquan District of Chengdu City participated in the survey. A total of 350 of these subjects were followed-up for 4 y after the initial cross-sectional survey,

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and 96.3% of the subjects participated in the complete data assessment in both the cross-sectional study and the follow-up survey. Finally, 240 subjects (91 men and 149 women) were included in our study. All of the subjects included were 18 to 70 y and had lived in Sichuan province for >5 y. The exclusion criteria included the following: 24 subjects had a history of arteriosclerosis, 46 exhibited carotid artery intima-media proliferation or plaque formation at baseline, as revealed by ultrasound examination, 6 were taking cholesterol-lowering medications, 27 were on blood pressure-lowering medications, 2 women were pregnant and 2 were cancer patients (Fig. 1).

This protocol was registered in the Chinese clinical trial registry under number #TR-CCH- ChiCTR-OCS-09000361. This study was revised and approved by The Ethics Committee of the China–Japan Friendship Hospital. Prior to the study, written informed consent was obtained from each individual. Blood samples were collected from subjects who were free of carotid artery intima-media proliferation and plaque formation between 2007 and 2008, and the plasma glucose concentration and serum ferritin levels were measured. Based on their plasma glucose concentrations, we classified the subjects into a normal glucose metabolism group and an abnormal glucose metabolism group, which included type 2 diabetes (T2DM), impaired fasting glucose (IFG) and impaired glucose tolerance (IGT), according to the diagnosis criteria of 1999 World Health Organization (WHO) diagnosis criteria [10]. Carotid intima-media thickness and carotid plaque were evaluated again using carotid artery color Doppler ultrasound during the follow-up in 2012.

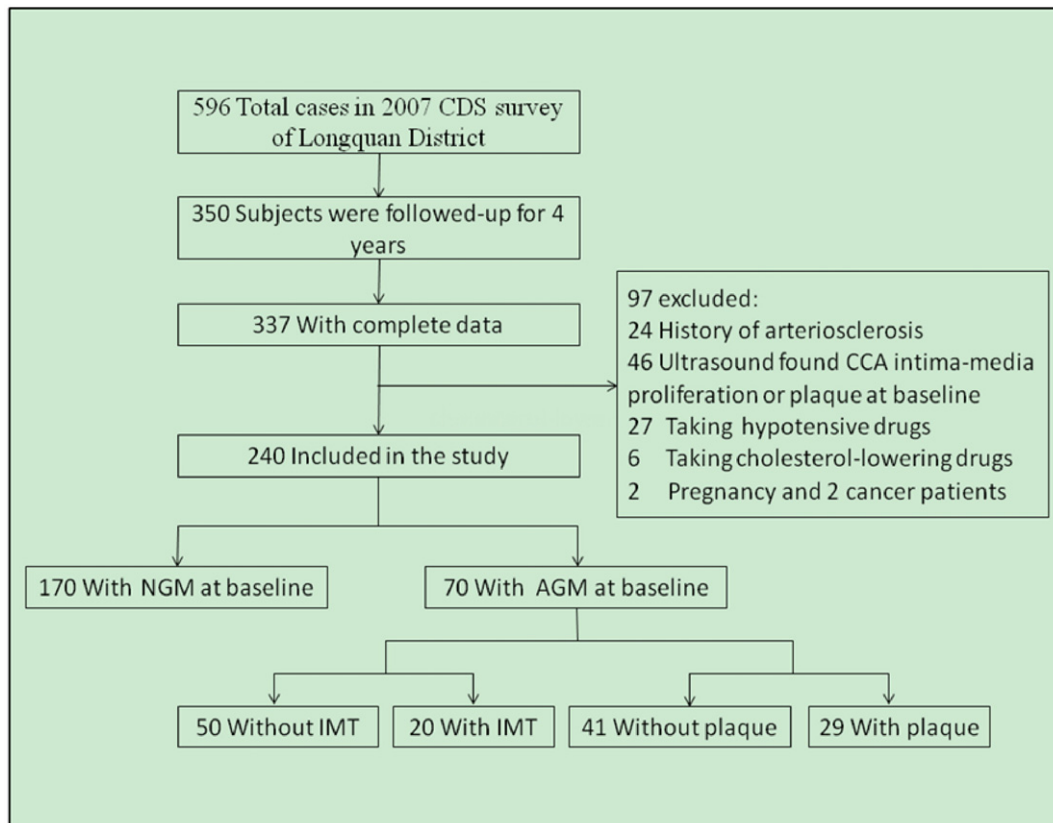
2.2. Carotid ultrasonography

Carotid artery lesions are described as common carotid artery intima-media thickening (IMT) or plaque formation. We used a high-

resolution B-mode ultrasound probe (IU22, Philips) equipped with a 7.5 MHz linear array to measure the common carotid artery (CCA). Participants were asked to remain in the supine position with a pillow under their shoulder during the examination. The subjects lifted their heads  $\leq 45^\circ$  and leaned slightly to the left for the measurement of the right CCA, and they then turned to the right for the measurement of the left CCA. The CCA was measured in multiple transverse and longitudinal planes along the 10 mm segment at the distal ends of the carotid bifurcation, and the mean internal diameter and the mean and maximum carotid intima-media thickness values for both sides were recorded. CCA IMT is characterized by a mean left and right CCA intima-media thickness  $\geq 1.0$  mm, whereas plaques are defined as local intima-media thickness  $\geq 1.3$  mm [11,12]. All of the images were evaluated and recorded by two sonographers who were blinded to the subjects' information and the test results.

2.3. Laboratory assessments

Trained staff recorded the lifestyle and clinical characteristics of the subjects with a standard questionnaire. A physical examination was performed on all subjects and included assessments of body height, weight, blood pressure, pulse, and waist and hip circumference. The subjects were consistent in their lifestyle, such as their diet and leisure-time physical activity, for at least 3 days before the administration of the oral glucose tolerance test (OGTT). After fasting for  $\geq 8$  h, venous blood samples were collected for the measurement of FPG measurement, fasting serum insulin (FINS), and other biochemical parameters. Blood samples were also obtained 120 min after the administration of a 75 g glucose load to measure plasma glucose and serum insulin levels. Plasma glucose levels were tested using a hexokinase enzymatic technique. A radioimmunoassay was employed to



CCA: common carotid artery; NGM: normal glucose metabolism; AGM: abnormal glucose metabolism; IMT: intima-media thickening.

Fig. 1. Study flow chart. CCA: common carotid artery; NGM: normal glucose metabolism; AGM: abnormal glucose metabolism; IMT: intima-media thickening.

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