Research Article

Fibrosis-4 Index Is Closely Associated with Arterial Damage and Future Risk of Coronary Heart Disease in Type 2 Diabetes

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This study evaluated the association between fibrosis-4 (FIB-4) index and arterial damage or future risk of coronary heart disease (CHD) in type 2 diabetes. The study subjects were 253 patients with type 2 diabetes. The FIB-4 index, as a marker of hepatic fibrosis based on age, aspartate aminotransferase and alanine aminotransferase levels, and platelet count, was calculated for all subjects. Carotid intima-media thickness (IMT), carotid artery calcification (CAC), and aortic arch calcification (AAC) grade (0–2) were assessed as atherosclerotic variables. The Suita score was calculated as the future risk of coronary heart disease (CHD). We assessed whether the FIB-4 index was associated with both atherosclerotic variables and the Suita score. FIB-4 index was significantly associated with IMT ($r = 0.241, P < 0.001$) and Suita score ($r = 0.291, P < 0.001$). Subjects with CAC showed a significantly higher FIB-4 index score compared to subjects without (1.70 ± 0.74 and 1.24 ± 0.69, respectively, $P < 0.001$), whereas the FIB-4 index was significantly elevated with a higher grade of AAC (1.24 ± 0.74, 1.56 ± 0.66, and 1.79 ± 0.71, respectively, $P < 0.001$). Linear regression analysis adjusted for clinical characteristics indicated that the FIB-4 index was positively associated with IMT, Suita score, CAC, and AAC grade ($\beta = 0.241, P = 0.004; \beta = 2.994, P < 0.001; \beta = 0.139, P = 0.001; \text{and } \beta = 0.265, P < 0.001$, respectively). FIB-4 index is closely associated with arterial damage and future risk of CHD in type 2 diabetes.

1. Introduction

Subjects with the traditional risk factors of cardiovascular disease (CVD), such as dyslipidemia, hypertension, diabetes mellitus, and smoking, have a high risk of developing CVD [1]. Particularly, patients with type 2 diabetes show a high risk of future CVD development [2]. Further, subjects who show a cluster of CVD risk factors, such as metabolic syndrome (MetS) and type 2 diabetes, have a high risk of CVD development [3]. Furthermore, it was reported that nonalcoholic fatty liver disease (NAFLD) and nonalcoholic liver steatohepatitis (NASH) are closely associated with CVD mortality [4, 5], whereas NAFLD and NASH are commonly related to MetS [6], especially, patients with type 2 diabetes show a high prevalence of NAFLD [7]. Indeed, patients with NAFLD show elevated Framingham Scores [8]. Thus, it is necessary to evaluate risk assessment for developing CVD to improve the prognosis of patients with NAFLD and NASH. NAFLD shows characteristic histology on liver biopsy, including steatosis, steatohepatitis, fibrosis, and cirrhosis [9], of which hepatic fibrosis is closely associated with mortality [10]. Since the development of glycemic abnormalities is correlated with an increased risk of hepatic fibrosis in Japanese subjects [11], it is necessary to evaluate whether hepatic fibrosis is closely related to atherosclerosis and future risk of CVD development in Japanese
patients with type 2 diabetes. Although several formulae using noninvasive variables exist to detect hepatic fibrosis, the fibrosis-4 (FIB4) index, which is a marker of hepatic fibrosis based on age, aspartate aminotransferase and alanine aminotransferase levels, and platelet count, is the most appropriate indicator of fibrosis in patients with NAFLD [12]. FIB4 index reflects the risk of hepatic fibrosis in type 2 diabetes. A previous study reported that the prevalence of hepatic fibrosis assessed via liver biopsy gradually increased, accompanied by an increase in the FIB 4 index in patients with type 2 diabetes who were originally diagnosed without or suspected of having hepatic fibrosis [13]. We concluded that the FIB4 index could be used to evaluate the risk of NAFLD and hepatic fibrosis in type 2 diabetes patients. Therefore, we hypothesized that the FIB4 index is associated with atherosclerosis and the risk of future CVD development.

We designed a cross-sectional study to determine the relationship between the FIB4 index and both atherosclerosis and future CVD risk in Japanese subjects with type 2 diabetes. In this study, we used carotid intima-media thickness (IMT) and aortic arterial calcification to evaluate arterial damage, and the Suita score of study subjects was calculated to estimate future coronary heart disease (CHD) risk. Carotid IMT [14] and aortic arch calcification (AAC) [15] are closely associated with future CVD risk, whereas the Suita score provides suitable risk factor categories for predicting the 10-year probability of CHD; it more accurately predicts CHD risk than the Framingham risk score in the Japanese population [16].

2. Materials and Methods

2.1. Study Subjects. Two hundred fifty-three patients with type 2 diabetes were enrolled in our study, comprised of 163 men and 90 women with an average age of 65.6 ± 10.9 years. All subjects were ambulatory and treated at either the Department of Neurology, Hematology, Metabolism, Endocrinology and Diabetology at the Yamagata University Faculty of Medicine or the Division of Diabetes and Metabolic Diseases, Department of Internal Medicine at Nihon University School of Medicine. All subjects had no history of hepatic cirrhosis, viral hepatitis, collagen diseases, including hepatic arteritis and primary biliary cirrhosis, autoimmune hepatitis, and malignant diseases. Patients with chest and cardiac diseases that affect the AAC evaluation, malignant diseases, collagen diseases, and acute and chronic inflammatory diseases and receiving immunosuppressants and steroid hormone therapy were excluded from our study.

2.2. Informed Consent and Ethics Regulations. The study was designed on the principles of the Declaration of Helsinki. We conducted a cross-sectional study using a clinical database. The study protocol was approved by the Ethics Committee of Yamagata University Faculty of Medicine and Nihon University School of Medicine. The protocol of this study is found on the web page of the Ethics Committee, and we provided the opportunity for participants to opt out of the study. The requirement for written informed consent was waived because of the retrospective nature of the study by our Ethics Committee.

2.3. Characteristics of Study Subjects. We evaluated the clinical characteristics of our study cohort, including sex, age, body mass index (BMI), smoking habit, systolic and diastolic blood pressure, hypertension, statin use, platelet (PLT) count, and biochemical variables. PLT and biochemical variables including lipid metabolism, uric acid, and HbA1c were measured after an overnight fast. PLT count was measured using a hematology analyzer, whereas low-density lipoprotein (LDL) and high-density lipoprotein (HDL) cholesterol, triglycerides, uric acid, creatinine, and HbA1c were measured using an automated analyzer. We used the estimated glomerular filtration rate (eGFR) as an indicator of renal function, which was determined by the following formula: eGFR (mL/min/1.73 m²) = 194 × serum creatinine⁻¹.094 × age⁻⁰.287 × 0.739 (if female) [17]. Blood pressure was measured in a sitting position at the hospital in the morning. Patients with hypertension were defined as patients treated with hypertension or patients with blood pressure greater than 140/90 mmHg that was measured in a hospital based on the Japanese Society of Hypertension Committee for Guidelines for the Management of Hypertension [18].

2.4. Examination of Hepatic Fibrosis, Atherosclerosis, and Cardiovascular Risk. The FIB4 index was calculated to evaluate hepatic fibrosis in our study subjects. It is a useful variable to assess hepatic fibrosis in Japanese patients with NAFLD [19], although the FIB4 index was originally established as a simple noninvasive index to predict significant fibrosis in a patient with human immunodeficiency virus (HIV)/hepatitis C virus (HCV) coinfection [12]. The FIB4 index was determined by the following formula [19]:

$$\text{FIB4 index} = \frac{\text{age (years)} \times \text{AST (IU/L)}}{\text{PLT count (10/L)} \times \sqrt{\text{ALT (IU/L)}}}$$

A FIB4 index with a cutoff of 1.45 provides suitable diagnostic accuracy to predict hepatic fibrosis in the Japanese population [19].

IMT was measured as a marker to evaluate arterial injury in our study subjects. Carotid IMT is a suitable surrogate marker for future CVD risk [14]. A total of six segments of the near and far walls in the common, bifurcation, and internal carotid artery on the right and left were measured by B-mode ultrasonography, as described in a previous study [20]. The maximum IMT, including a plaque on both sides, was defined as the IMT in all study subjects [20]. Trained physicians or ultrasonographers performed the carotid ultrasonographic measurements. IMT measurements showed variability of 8.0% in our cohort, as previously reported [20].

Carotid artery calcification (CAC) and the grade of AAC were evaluated as measures of arterial calcification. CAC and AAC are closely related to future CVD risk [15, 21]. The presence (n = 141) or absence (n = 112) of CAC on both sides
was recorded using B-mode ultrasonography. Poster- 
oanterior chest X-ray films were performed for study 
subjects, and AAC grade was reviewed in a manner as previously 
reported [22, 23]. Subjects were divided into three groups 
based on the grade of AAC as follows: grade 0, no visible 
calcification (n = 95, Figure 1(a) online); grade 1, small spots 
or a single thin area of calcification (n = 73, Figure 1(b) 
online); or grade 2, one or more areas of thick calcification or 
circumferential calcification (n = 83, Figures 1(c) and 1(d)). 
AAC assessment in this manner shows good accuracy and reproducibility [22]. In our study, one observer performed an AAC assessment. An evaluation of AAC grade showed reproducibility of 90.0% [22].

The Suita score was estimated to predict the risk of future 
CHD in study subjects. It is an established cardiovascular 
risk score using risk factor categories for predicting CHD in 
the Japanese population based on age, gender, smoking, 
diabetes, hypertension stage, LDL and HDL cholesterol, and 
chronic kidney disease (CKD) stage [16]. The Suita score 
consists of the sum of each risk score and indicates the 10-
year probability of CHD [16].

2.5. Statistical Analysis. Pearson’s coefficient correlation and 
univariate linear regression analysis were used to identify 
whether clinical characteristics, IMT, and Suita score were 
significantly associated with the FIB4 index. In this un-
ivariate analysis, we assumed the FIB4 index as a dependent 
variable and sex (men), smoking habit (current), hyperten-
sion, and statin use as independent variables. The Mann–
Whitney U test was performed to compare the mean 
value of the FIB4 index in subjects between the presence 
and absence of CAC. Bonferroni’s multiple comparison test was 
used to compare the mean value of the FIB4 index between 
AAC grades. Univariate and multivariate linear regression 
analysis adjusted for clinical characteristics was performed 
to certify whether IMT and Suita scores were significantly 
associated with the FIB4 index. In this univariate and 
multivariate analysis, the FIB4 index was assumed as an 
independent variable, and IMT, Suita score, CAC, and AAC 
grade were assumed as dependent variables. Data are 
presented as means ± standard deviation (SD), number (%), 
coefﬁcient covariation (r), or β coefficients. Statistical signif-
ificance was defined as P < 0.05. All analyses were per-
formed with IBM SPSS Statistics for Windows Version 25 J 
(IBM Corp., Armonk, NY, USA).

3. Results

3.1. Characteristics of Study Subjects. Table 1 shows the 
clinical characteristics of our study cohort. A total of 163 
subjects (64.4%) were men with an overall mean age of 
65.6 ± 10.9 years. The number of patients with hypertension 
and receiving statins was 195 (77.1%) and 147 (58.1%), re-
spectively. The mean values of systolic and diastolic blood 
pressure, HDL and LDL cholesterol, and triglycerides were 
130.6 ± 16.8 and 76.4 ± 11.2 mmHg and 1.52 ± 0.74, 1.41 ± 0.45, 
and 1.55 ± 0.98 mmol/L, respectively. The mean values of BMI, 
eGFR, and HbA1c were 25.09 ± 4.52, 74.14 ± 25.16 mL/min/ 
1.73 m², and 7.95 ± 1.88%, respectively.

3.2. Relationships between FIB4 Index and Clinical Charac-
teristics of Study Subjects. Additionally, we found that the 
mean FIB4 index was 1.52 ± 0.74 (Table 1), and the number of 
patients with no risk of hepatic fibrosis (FIB4 index ≤1.45) 
was 130 (51.4%; 82 men and 48 women). Diastolic blood 
pressure (r = −0.189, P < 0.001), eGFR (r = −0.243, P < 0.001), 
and HbA1c (r = −0.206, P = 0.001) were significantly in-
versely correlated with the FIB4 index in our cohort (Table 2).

3.3. Relationships between FIB4 Index and Both Atheroscle-
rosis and Future Risk of CVD. Positive correlations were 
found between the FIB4 index and IMT and Suita score 
(r = 0.241 and 0.291, respectively, both P < 0.001, Figure 2). 
Regarding associations between the FIB4 index and arterial 
calcification, we found that the FIB4 index in subjects with 
CAC (n = 141) was significantly higher than that in subjects 
without (n = 112) (1.70 ± 0.69 and 1.24 ± 0.74, respectively, 
P < 0.001, Figure 3). Furthermore, the FIB4 index of patients with 
AAC grade 1 (n = 73) or 2 (n = 83) was significantly higher 
than that found in patients with grade 0 (n = 95) (P = 0.001 
and P < 0.001, respectively, Figure 3), although the FIB4 
index increased according to a higher grade of AAC 
(1.24 ± 0.74, 1.56 ± 0.66, and 1.79 ± 0.71, respectively, Fig-
ure 3). Linear regression analysis indicated that the FIB4 
index indicated significantly positive associations among 
IMT, Suita score, CAC, and AAC grade (β = 0.312, 3.573, 
0.183, and 0.355, respectively, P < 0.001) (Table 3). Similarly, 
linear regression analysis adjusted for diastolic blood 
pressure, eGFR, and HbA1c indicated that the FIB4 index 
showed significant positive associations between IMT 
(β = 0.241, P = 0.004), Suita score (β = 2.994, P < 0.001), 
CAC (β = 0.139, P = 0.001), and AAC grade (β = 0.265, 
P < 0.001) (Table 3).

4. Discussion

In this study, we found that the FIB4 index was closely 
associated with arterial damage, such as carotid intimal 
thickening and arterial calcification in two major vessel beds 
(carotid and aortic arch). In particular, the FIB4 index was 
significantly increased according to the higher grade of 
calcification in the aortic arch. Further, we found a signif-
icantly positive correlation between the FIB4 index and the 
Suita score, which is a predictive marker of future CHD 
development. The results of this study support the new 
insight that noninvasive hepatic fibrosis score is significantly 
related to arterial damage and future risk of CHD in Jap-
anese patients with type 2 diabetes. The clinical utility of 
the results of our study is that arterial damage can be easily 
estimated using some biochemical data (PLT count, AST, 
and ALT) and age in type 2 diabetes patients. Evaluation of 
arterial damage in patients using computed tomography, 
magnetic resonance imaging, carotid artery ultrasonogra-
phy, pulse wave velocity, and endothelial function, among 
others, cannot be frequently performed. In contrast, the 
FIB4 index can be calculated instantly using the results of
blood sampling in general practice or medical checkups. As a result, physicians can easily and frequently estimate arterial damage in patients with type 2 diabetes during general consultations or medical checkups using the FIB4 index. We conclude that the FIB4 index may be a useful tool for identifying patients with type 2 diabetes who are at high risk of CHD.

The effect of NAFLD and NASH on the future risk of CVD and mortality remains unclear. A meta-analysis study reported that cardiovascular mortality was not significantly related to NAFLD, although NAFLD increased the risk of cardiovascular events [24]. However, more severe NAFLD was significantly associated with cardiovascular mortality [24]. Indeed, some retrospective cohort studies showed that patients with NASH, but not those with simple steatosis, have a higher risk of CVD mortality [4, 10, 25]. Thus, the results of these studies suggest that hepatic fibrotic change in NAFLD and NASH is more closely related to future CVD risk and mortality than steatosis. Indeed, recent studies have shown that noninvasive scores of hepatic fibrosis were positively correlated with cardiovascular risk scores in NAFLD patients [26].

We considered some reasons why hepatic fibrosis becomes a risk of arterial damage and future CVD based on the results of past studies. First, we considered whether the worsening of glucose metabolism accompanied by the progression of hepatic fibrosis had an important role. Interestingly, overall mortality and death from CVD in patients with NAFLD without diabetes were not significantly elevated compared to those in control patients [4]. Therefore, impaired glucose metabolism may elevate the risk of CVD mortality in patients with hepatic fibrosis. A study of patients with liver cirrhosis (LC) found that insulin resistance in both the periphery (skeletal and adipose) and liver was independent of LC severity [6]. Additionally, patients with LC and diabetes showed hyperglycemia and hyperinsulinemia after a 75g glucose load compared with patients with either normal or impaired glucose tolerance [6]. These results indicate that postprandial hyperglycemia and hyperinsulinemia induced by insulin resistance have an important role in the development of both arterial damage and CVD in patients with hepatic fibrosis. Postprandial hyperglycemia elevates arterial stiffness [27], induces carotid IMT thickening [28], and increases the risk of CVD development [29]. Hyperinsulinemia also elevates arterial stiffness [30], induces arterial intimal thickening [31], and raises CVD mortality [32].

Second, we conclude that systematic metabolic abnormalities in patients with NAFLD and NASH also contribute to the progression of arterial damage and CVD. NAFLD-induced production of pathological factors promotes multiple aspects of atherosclerosis progression. Increase of lipid

![Figure 1: Classification of aortic arch calcification (AAC) grade. (a) grade 0, (b) grade 1, and (c, d) grade 2.](image)

| Table 1: Clinical characteristics of study subjects. |
|---------------------------------------------------|
| Clinical characteristics | Mean ± SD, n (%) | Range |
| Sex (men) | 163 (64.4) | |
| Age (years) | 65.6 ± 10.9 | 35–86 |
| Body mass index | 25.09 ± 4.52 | 14.3–40.1 |
| Smoking habit (current) | 55 (21.7) | |
| Systolic blood pressure (mmHg) | 130.6 ± 16.8 | 92–181 |
| Diastolic blood pressure (mmHg) | 76.4 ± 11.2 | 43–105 |
| Hypertension | 195 (77.1) | |
| Statin use | 147 (58.1) | |
| PLT (×10^9/L) | 231.9 ± 60.7 | 111–531 |
| ALT (IU/L) | 24.8 ± 14.3 | 7–143 |
| AST (IU/L) | 26.7 ± 21.6 | 7–232 |
| FIB4 index | 1.52 ± 0.74 | 0.36–4.84 |
| HDL-CHOL (mmol/L) | 1.41 ± 0.45 | 0.65–3.81 |
| LDL-CHOL (mmol/L) | 2.66 ± 0.84 | 0.85–8.86 |
| Triglyceride (mmol/L) | 1.55 ± 0.98 | 0.36–7.91 |
| Uric acid (µmol/L) | 325.5 ± 77.8 | 59.5–630.5 |
| eGFR (mL/min/1.73 m²) | 74.14 ± 25.16 | 14.9–176.9 |
| HbA1c (%) | 7.95 ± 1.88 | 5.6–18.3 |

SD, standard deviation; PLT, platelet; ALT, alanine aminotransferase; AST, aspartate aminotransferase; FIB4 index, fibrosis-4 index; HDL-CHOL, high-density lipoprotein cholesterol; LDL-CHOL, low-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; HbA1c, glycosylated hemoglobin.
Table 2: Associations between clinical characteristics and FIB4 index.

| Clinical characteristics          | Correlation coefficient | β-coefficient (95% CI)          | P value |
|----------------------------------|-------------------------|---------------------------------|---------|
| Sex (men)                        | -0.097                  | -0.049 (-0.242–0.143)           | 0.616   |
| Body mass index                  |                         |                                 | 0.122   |
| Smoking habit (current)          | -0.099                  | -0.099 (-0.220–0.021)           | 0.105   |
| Systolic blood pressure (mmHg)   | 0.051                   |                                 | 0.417   |
| Diastolic blood pressure (mmHg)  | -0.189                  |                                 | 0.003   |
| Hypertension                     |                         | 0.201 (-0.017–0.419)           | 0.071   |
| Statin use                       |                         | 0.044 (-0.143–0.231)           | 0.642   |
| HDL-CHOL (mmol/L)                | 0.095                   |                                 | 0.131   |
| LDL-CHOL (mmol/L)                | -0.060                  |                                 | 0.341   |
| Triglyceride (mmol/L)            | 0.001                   |                                 | 0.990   |
| Uric acid (μmol/L)               | 0.052                   |                                 | 0.413   |
| eGFR (mL/min/1.73 m²)            | -0.243                  |                                 | <0.001  |
| HbA1c (%)                        |                         | -0.206                          | 0.001   |

FIB4 index, fibrosis-4 index; HDL-CHOL, high-density lipoprotein cholesterol; LDL-CHOL, low-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; HbA1c, glycosylated hemoglobin; CI, confidence interval.

Figure 2: Correlations between FIB4 index and both (a) IMT and (b) Suita score. FIB4 index, fibrosis-4 index; IMT, intima-media thickness.

Figure 3: Distribution of FIB4 index in (a) CAC and (b) AAC grade. ** P < 0.001 vs. without; * P = 0.001 and ** P < 0.001 vs. AAC grade 0. FIB4 index, fibrosis-4 index; CAC, carotid artery calcification; AAC, aortic arch calcification.
The FIB4 index shows a positive association with age (the numerator of the FIB4 index formula contains age, the blood pressure in our study account for the significant in-associations between age and both HbA1c and diastolic by AAC progression in type 2 diabetes [23].

Interestingly, we demonstrated HbA1c and diastolic blood pressure to be inversely associated with the FIB4 index. The reasons for these inverse associations remain unknown. We conclude that there may be a negative relationship between the FIB4 index and HbA1c or diastolic blood pressure. We believe that younger subjects in our study had a more difficult time improving their glycemic control. Indeed, our study subjects who received insulin therapy were younger and had relatively high HbA1c levels (data not shown). Furthermore, diastolic blood pressure is generally decreased according to aging [43].

There were several possible limitations in our study. First, we calculated the FIB4 index in subjects including patients at low risk of NAFLD/NASH. In principle, the FIB4 index is recommended for evaluating hepatic fibrosis in patients with NAFLD/NASH, not in patients without NAFLD/NASH. We considered that the FIB4 index was linked to the risk of hepatic fibrosis because the prevalence of actual hepatic fibrosis in patients with type 2 diabetes rose in tandem with an increase in the FIB4 index underlying liver disease [13]. As a result, evidence that the FIB4 index is significantly associated with arterial damage and future risk of CHD in subgroup type 2 patients with or without NAFLD/NASH is required. However, we do not have any data on hepatic imaging, such as computed tomography, magnetic resonance imaging, or ultrasonography. Therefore, we were unable to analyze the relationship between the FIB4 index and CHD risk in the subgroups with and without NAFLD. We aim to clarify these relationships in future research. Second, study subjects may include patients with undiagnosed chronic hepatic disease, although subjects who had no history of hepatic cirrhosis, viral hepatitis, collagen diseases, including hepatic arteritis and primary biliary cirrhosis, autoimmune hepatitis, and malignant diseases were excluded. Finally, the reproducibility of the FIB4 index was not shown. We believe our study results should be validated in patients with high reproducibility in the FIB4 index at different times to increase the clinical value of our study results.

5. Conclusions

This study provides evidence that noninvasive assessment of hepatic fibrosis using the FIB4 index is closely associated with arterial damage and future risk of CHD in Japanese patients with type 2 diabetes. The results of our study also indicate that the FIB4 index is a suitable tool to identify those patients with diabetes at a higher risk of CVD.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this article.

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