Association between non-alcoholic fatty liver disease evaluated by transient elastography with extracranial carotid atherosclerosis in a multiethnic Asian community

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Key words
atherosclerosis, carotid intima-media thickness, Fibroscan, non-alcoholic fatty liver disease, transient Elastography, ultrasound carotid Doppler.

Abstract

Background and Aim: There is not much data on the association between non-alcoholic fatty liver disease (NAFLD) and advanced fibrosis assessed using Fibroscan with carotid intima-media thickness (CIMT) in the general population. The objective of this study was to evaluate the association between NAFLD and advanced fibrosis, as diagnosed by Fibroscan, with an increased CIMT in the Malaysian population.

Methods: A cross-sectional study of government officers and their family members attending a health screening at a public healthcare facility was conducted. All subjects underwent clinical evaluation, biochemical testing, anthropometry, ultrasound carotid Doppler, and Fibroscan examination.

Results: Data for 251 subjects were analyzed (mean age 47.1 ± 12.4 years, 74.1% male). Prevalence of NAFLD and advanced fibrosis were 57.4 and 17.5%, respectively. Independent factors associated with NAFLD were waist circumference (odds ratio [OR] = 1.077, 95% confidence interval [CI] 1.038–1.118, P < 0.001) and serum alanine aminotransferase (ALT) (OR = 1.039, 95% CI 1.005–1.074, P = 0.024). Independent factors associated with advanced fibrosis were male gender (OR = 4.847, 95% CI 1.369–17.155, P = 0.014) and serum aspartate aminotransferase (AST) (OR = 1.057, 95% CI 1.003–1.113, P = 0.036). Prevalence of increased CIMT was 29.0%. Independent factor associated with increased CIMT was older age (OR = 1.146, 95% CI 1.067–1.231, P < 0.001). Of the subjects, 34.5% with NAFLD had increased CIMT compared to 19.1% of the subjects without NAFLD (P = 0.063). Advanced fibrosis was not associated with increased CIMT.

Conclusions: Prevalence of NAFLD, advanced liver fibrosis, and increased CIMT were high. NAFLD and advanced liver fibrosis appeared not to be associated with increased CIMT. However, a larger sample size is needed to demonstrate whether there is any association.

Introduction

The prevalence of non-alcoholic fatty liver disease (NAFLD) is rising all over the world.1 Non-alcoholic steatohepatitis (NASH), the more severe form of NAFLD, can progress to fibrosis and cirrhosis. NASH is a leading cause of liver transplantation due to hepatic failure and liver cancer.2 Recent noninvasive modalities, such as liver stiffness measurement (LSM) with transient elastography, have gradually replaced liver biopsy for the estimation of liver fibrosis.3 Controlled attenuation parameter (CAP), which measures the decrease in amplitude of ultrasound as it transmits through liver tissue, is excellent for the evaluation of significant hepatic steatosis. CAP is estimated using the same radiofrequency data used for the assessment of LSM with Fibroscan, an ultrasound-based vibration-controlled transient elastography device.4 Carotid intima-media thickness (CIMT) is used to detect subclinical atherosclerosis.5 Progression of CIMT leads to plaque development, carotid stenosis and, subsequently, a higher risk of stroke.5 In a previous cross-sectional study, NAFLD evaluated by liver ultrasonography was found to be associated with an increased CIMT.6 A systematic review reported that 13% of NAFLD patients, diagnosed through simple transabdominal ultrasound scanning, have an increased CIMT.7 To date, there is only one study on the prevalence of NAFLD in the general population in Malaysia.8 So far, there has been no report on the relationship of NAFLD, as evaluated by Fibroscan, with CIMT. The primary objective of this study was to evaluate the association between...
NAFLD, as diagnosed by Fibroscan, with an increased CIMT in the Malaysian population. The secondary objective was to assess the association between advanced liver fibrosis with increased CIMT. We also evaluated the risk factors associated with NAFLD, advanced fibrosis, and increased CIMT.

**Methods**

Ethics approval was obtained from University Malaya Medical Centre (MREC ID No.: 201410-677) before the study was conducted. The study was a cross-sectional study on consecutive government officers and their family members who attended a health screening at a public healthcare facility from August 2015 to January 2016. Study participants with significant alcohol intake or those known to have human immunodeficiency virus (HIV) infection, chronic hepatitis B virus infection, chronic hepatitis C virus infection, or other chronic liver diseases were excluded from participating in this study. Pregnant women were also not included in this study.

Demographic, anthropometric, clinical, and laboratory data were obtained using standard protocol. The intake of alcohol was estimated with the quantity–frequency method.9 Significant alcohol intake was defined as ≥21 units per week for men and ≥14 units per week for women. The subject’s height and weight was measured using standardized equipment. Body mass index (BMI) was calculated by dividing weight (in kilogram) by the square of height (in meters). Obesity was defined as BMI ≥25.0 kg per m². Waist circumference was measured at the mid-point between the lowest margin of the least palpable rib and the top of the iliac crest, and this was measured in the standing position. Central obesity was defined as waist circumference >90 cm in the men and >80 cm in the women.10 Blood pressure was measured in a sitting position using standard electronic blood pressure measuring equipment. The subject was defined as having hypertension if he or she was on antihypertensive medication or had systolic blood pressure (SBP) of ≥140 mmHg and diastolic blood pressure (DBP) ≥90 mmHg. The Global Physical Activity Questionnaire, which was developed by the World Health Organization, was used to measure the physical activity in three domains, namely, activity at work, travel to-and-from places, and recreational activities.11

Blood was taken from all the subjects after an overnight fast for full blood count, liver profile, fasting lipid profile, hepatitis B and C, and HIV screening. An oral glucose tolerance test was performed for all subjects. A subject was considered to have dyslipidemia if he or she was on lipid-lowering medication or if the subject had the serum total cholesterol (TC) ≥5.2 mmol/L, serum low-density lipoprotein (LDL) ≥3.4 mmol/L, serum high-density lipoprotein (HDL) <1.0 mmol/L in men or <1.3 mmol/L in women, or serum triglyceride (TG) ≥1.7 mmol/L. A subject was considered to have type 2 diabetes mellitus if he or she was on antidiabetic medication or if the subject had a fasting blood sugar (FBS) of ≥7.0 mmol/L and the 2-h postprandial blood sugar of ≥11.1 mmol/L. A subject was considered to have the metabolic syndrome if three or more of the following were present: hypertension; central obesity; hypertriglyceridemia; low serum HDL; and impaired fasting glucose, impaired glucose tolerance, or diabetes mellitus.10

**Fibroscan examination.** Fibroscan examination was performed using FibroScan® 502 Touch with M-probe (EchoSens, Paris, France) by a certified operator blinded to the clinical data and the results of Doppler ultrasonography of the carotid artery. An examination was successful if 10 valid measurements were obtained and reliable if the interquartile range (IQR)/median was ≤30%. An examination could still be considered reliable when the IQR/median was ≤30% if the LSM was <7.1 kPa.12 Subjects with unreliable examination were not included in the data analysis. Advanced fibrosis was diagnosed based on LSM ≥28 kPa.3,13 Significant hepatic steatosis was diagnosed based on CAP measurement ≥263 dB/m.4

**Ultrasound carotid Doppler.** Ultrasound carotid Doppler was performed using LOGIQ e (General Electric Healthcare, Chicago, IL, USA) with a 12 MHz probe by an experienced operator blinded to clinical data and Fibroscan examination results. The examination was performed according to standard protocol. CIMT measurements were taken at three different angles from the distal 1 cm of the far wall of both common carotid arteries. An average value was taken for both right and left common carotid arteries. An increased CIMT was defined as CIMT ≥0.8 mm.14

**Statistical analysis.** With an estimated prevalence of 20%, (11) the formula \( n = Z^2 P (1 - P)/d^2 \) was used for calculation of the sample size \( n \) = sample size, \( Z \) = Z statistic for a level of confidence, \( P \) = proportion \( [P ≥ 0.2] \) and \( d \) = precision \([d = 0.05])\). A total of 246 subjects were required to estimate the prevalence with 95% confidence. Statistical analysis was performed using Statistical Package for Social Sciences (SPSS) version 16.0 (SPSS Inc., Chicago, USA). Categorical variables were expressed as percentages and analyzed using Pearson Chi-square test or Fisher’s exact test where appropriate. Continuous variables were analyzed using Student’s t-test or Mann Whitney U test where appropriate. Continuous variables were expressed as means ± standard deviation or median with interquartile range. Factors associated with NAFLD, advanced fibrosis, and increased CIMT were then analyzed using univariate and multiple logistic regression analysis. A \( P \) value of <0.05 was considered statistically significant. If the categorical and continuous forms of the variable were both found to be statistically significant, then the categorical form of the variable was chosen for multiple logistic regression analysis.

**Results**

**Study population.** Of the 356 subjects who attended the health screening during the study period, 332 subjects consented to participate in the study; 81 subjects were excluded from the study (57 incomplete data, 8 significant alcohol intake, 2 chronic hepatitis B infection, 6 failed Transient Elastography (TE), 8 had unreliable TE). Therefore, 251 subjects were included in the data analysis (Fig. 1). The mean age of the study population was 47.1 ± 12.4 years old, consisting of 74.1% men. Obesity and central obesity were observed in 64.5 and 74.5%, respectively. Hypertension, diabetes mellitus, dyslipidemia, and the metabolic syndrome were observed in 30.3, 9.6, 76.1, and 40.2%, respectively. The study population consisted of 68.1% Malay, 19.9% Chinese, and 33.1% Indians. The prevalence of NAFLD, advanced fibrosis, and increased CIMT in the overall population and according to the different ethnic groups are shown in Table 1.

**Prevalence of NAFLD and the associated factors.** The prevalence of NAFLD in the overall population was 57.4%
Table 2 shows the characteristics of subjects with and without NAFLD. The subjects with NAFLD were significantly older in age. They were also significantly more likely to be male and to have obesity, central obesity, and hypertension. They were also more likely to have elevated SBP, DBP, serum TC, TG, alanine aminotransferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyl transferase (GGT) levels. A total of 51.4% (74/144) subjects with NAFLD had metabolic syndrome, whereas 25.2% (27/107) subjects without NAFLD had metabolic syndrome (P < 0.001). On multivariate analysis, the independent factors that were associated with NAFLD were waist circumference (odds ratio [OR] = 1.077, 95% confidence interval [CI] 1.038–1.118, P < 0.001) and serum ALT (OR = 1.039, 95% CI 1.005–1.074, P = 0.024) (Table 3).

Prevalence of advanced fibrosis and the associated factors. The prevalence of advanced liver fibrosis in the overall population was 17.5% (44/251). Table 4 shows the characteristics of subjects with and without advanced fibrosis. The subjects with advanced fibrosis were significantly more likely to be male and were more likely to have dyslipidemia. They had higher BMI; waist circumference SBP; and serum TC, ALT, AST, and GGT levels. A total of 59.1% (26/44) subjects with advanced fibrosis had metabolic syndrome, whereas 36.2% (75/207) without advanced fibrosis had metabolic syndrome (P = 0.007). On multivariate analysis, the independent factors associated with advanced fibrosis were serum AST (OR = 1.057, 95% CI 1.003–1.113, P = 0.036) and male gender (OR = 4.847, 95% CI 1.369–17.155, P = 0.014) (Table 5).

Prevalence of increased CIMT and associated factors. Over half of the study population (131/251, 52.2%) underwent ultrasound carotid Doppler. Increased CIMT was observed in 29.0% (38/131). Table 6 shows the characteristics of subjects with and without increased CIMT. Subjects with increased CIMT were significantly older, and they were more likely males. They

| Table 1 | Prevalence of NAFLD, advanced fibrosis, and increased CIMT in the various ethnic groups |
|------------------|------------------|------------------|------------------|------------------|
|                 | Overall population | Malay            | Chinese          | Indians          |
| Prevalence of NAFLD, % (n/N) | 57.4 (144/251) | 56.1 (96/171) | 17 (29/171) | 30 (19/62) |
| Prevalence of advanced fibrosis, % (n/N) | 17.5 (44/251) | 17 (29/171) | 23.3 (7/30) | 16 (8/50) |
| Prevalence of raised CIMT, % (n/N) | 29 (38/131) | 25 (19/76) | 30 (6/20) | 37.1 (13/35) |

P values are those comparing the prevalence of NAFLD, advanced fibrosis, and raised CIMT across the different ethnic groups. CIMT, carotid intima-media thickness; NAFLD, non-alcoholic fatty liver disease.
were also more likely to have diabetes mellitus and hypertension. They had greater waist circumference and higher SBP, FBS, LDL, and ALT. A total of 50% (19/38) of the subjects with increased CIMT had metabolic syndrome, whereas 40.8% (49/93) subjects without increased CIMT had metabolic syndrome ($P = 0.17$); 34.5% (29/84) of the subjects with NAFLD had an increased CIMT compared to 19.1% (9/47) of the subjects without NAFLD ($P = 0.063$); 30.4% (7/23) of the subjects with advanced fibrosis had an increased CIMT compared to 28.7% (31/108) of the subjects without advanced fibrosis ($P = 0.868$).

On multivariate analysis, the only independent factor associated with increased CIMT was older age (OR = 1.146, 95% CI 1.067–1.231, $P < 0.001$) (Table 7).

**Discussion**

This study has demonstrated several important observations on NAFLD and CIMT in a community-based setting in Malaysia. We observed an alarmingly high prevalence of NAFLD (57.4%). Malaysia is one of the countries with the highest prevalence of

**Table 2** Characteristics of participants with and without NAFLD

|                              | Participants with NAFLD, $n = 144$ | Participants without NAFLD, $n = 107$ | $P$ value |
|------------------------------|------------------------------------|---------------------------------------|-----------|
| Age, years                   | 49.1 ± 11.0                        | 44.4 ± 13.5                           | 0.002     |
| Gender, $n$ (%)              |                                    |                                       |           |
| Male                         | 121 (84)                           | 65 (60.7)                             | <0.0001   |
| Female                       | 23 (16)                            | 42 (39.3)                             |           |
| Ethnic group, $n$ (%)        |                                    |                                       |           |
| Malay                        | 96 (66.7)                          | 75 (70.1)                             | 0.280     |
| Chinese                      | 15 (10.4)                          | 15 (14.0)                             |           |
| Indian                       | 33 (22.9)                          | 17 (15.9)                             |           |
| Smoking, $n$ (%)             |                                    |                                       |           |
| Yes                          | 33 (22.9)                          | 18 (16.8)                             | 0.235     |
| No                           | 111 (77.1)                         | 89 (83.2)                             |           |
| Diabetes mellitus, $n$ (%)   |                                    |                                       |           |
| Yes                          | 13 (9)                             | 11 (10.3)                             | 0.739     |
| No                           | 131 (91)                           | 96 (89.7)                             |           |
| Hypertension, $n$ (%)        |                                    |                                       |           |
| Yes                          | 55 (38.2)                          | 21 (19.6)                             | 0.002     |
| No                           | 89 (61.8)                          | 86 (80.4)                             |           |
| Dyslipidemia, $n$ (%)        |                                    |                                       |           |
| Yes                          | 116 (80.6)                         | 75 (70.1)                             | 0.055     |
| No                           | 28 (19.4)                          | 32 (29.9)                             |           |
| BMI, kg per m$^2$            | 27.7 ± 3.7                         | 24.7 ± 3.4                            | <0.0001   |
| Obesity, $n$ (%)             |                                    |                                       |           |
| Yes                          | 110 (76.4)                         | 52 (48.6)                             | <0.0001   |
| No                           | 34 (23.6)                          | 55 (51.4)                             |           |
| Waist circumference, cm      | 96.9 ± 9.1                         | 88.6 ± 9.4                            | <0.0001   |
| Central obesity, $n$ (%)     |                                    |                                       |           |
| Yes                          | 125 (86.8)                         | 62 (57.9)                             | <0.0001   |
| No                           | 19 (13.2)                          | 45 (42.1)                             |           |
| SBP, mmHg                    | 138 ± 14                           | 128 ± 13                              | <0.0001   |
| DBP, mmHg                    | 83 ± 10                            | 76 ± 11                               | <0.0001   |
| FBS, mmol/L                  | 5.5 ± 1.2                          | 5.3 ± 1.4                             | 0.283     |
| TC, mmol/L                   | 5.7 ± 1.0                          | 5.4 ± 1.2                             | 0.049     |
| HDL, mmol/L                  | 1.4 ± 0.3                          | 1.5 ± 0.3                             | 0.049     |
| LDL, mmol/L                  | 3.1 ± 1.2                          | 2.9 ± 1.2                             | 0.087     |
| TG, mmol/L                   | 1.8 ± 1.1                          | 1.5 ± 0.6                             | 0.017     |
| ALT, IU/L                    | 38 (28–43)                         | 28 (21–36)                            | <0.0001   |
| AST, IU/L                    | 28 (21–38)                         | 24 (19–32)                            | 0.003     |
| GGT, IU/L                    | 54 (31–65)                         | 38 (22–58)                            | <0.0001   |

Alt, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; DBP, diastolic blood pressure; FBS, fasting blood sugar; GGT, gamma-glutamyl transferase; GPAQ, Global Physical Activity Questionnaire; HDL, high-density lipoprotein; LDL, low-density lipoprotein; NAFLD, non-alcoholic fatty liver disease; SBP, systolic blood pressure; TC, total cholesterol; TG, triglyceride; WHO, World Health Organization.
### Table 3: Univariate and multivariate analysis of factors associated with NAFLD

|                      | Univariate analysis |                      |                      |                      | Multivariate analysis |                      |                      |                      |
|----------------------|---------------------|----------------------|----------------------|----------------------|-----------------------|----------------------|----------------------|----------------------|
|                      | β                   | OR                   | 95% CI               | P value              | β                     | Adjusted OR         | 95% CI               | P value              |
| Age                  | 0.032               | 1.033                | 1.011–1.055          | 0.003                | 0.021                 | 1.021               | 0.995–1.048          | 0.114                |
| Male                 | 1.224               | 3.399                | 1.882–6.139          | <0.001               | 0.531                 | 1.701               | 0.861–3.361          | 0.126                |
| Waist circumference  | 0.102               | 1.108                | 1.070–1.146          | <0.001               | 0.074                 | 1.077               | 1.038–1.118          | >0.001               |
| ALT                  | 0.050               | 1.052                | 1.028–1.075          | <0.001               | 0.038                 | 1.039               | 1.005–1.074          | 0.024                |
| AST                  | 0.045               | 1.046                | 1.016–1.077          | 0.002                | –0.005                | 0.995               | 0.949–1.042          | 0.819                |
| GGT                  | 0.020               | 1.020                | 1.008–1.032          | 0.001                | 0.004                 | 1.004               | 0.989–1.019          | 0.628                |
| Dyslipidemia         | 0.570               | 1.768                | 0.985–3.171          | 0.056                | 0.129                 | 1.138               | 0.579–2.336          | 0.707                |
| Hypertension         | 0.929               | 2.531                | 1.412–4.536          | 0.002                | 0.354                 | 1.425               | 0.713–2.848          | 0.316                |

ALT, alanine aminotransferase; AST, aspartate aminotransferase; CI, confidence interval; GGT, gamma-glutamyl transferase; NAFLD, non-alcoholic fatty liver disease; OR, odds ratio.

### Table 4: Characteristics of participants with and without advanced liver fibrosis

|                      | Participants with advanced fibrosis, n = 44 | Participants without advanced fibrosis, n = 207 | P value |
|----------------------|--------------------------------------------|-------------------------------------------------|---------|
| Age, years           | 50.2 ± 12.9                                | 46.4 ± 12.2                                     | 0.083   |
| Gender, n (%)        |                                            |                                                 |         |
| Male                 | 41 (93.2)                                  | 145 (70)                                       | 0.001   |
| Female               | 3 (6.8)                                    | 62 (30)                                        |         |
| Ethnic group, n (%)  |                                            |                                                 |         |
| Malay                | 29 (65.9)                                  | 142 (68.6)                                     | 0.827   |
| Chinese              | 7 (15.9)                                   | 23 (11.1)                                      |         |
| Indian               | 8 (18.2)                                   | 42 (20.3)                                      |         |
| Smoking, n (%)       |                                            |                                                 |         |
| Yes                  | 12 (27.3)                                  | 39 (18.8)                                      | 0.207   |
| No                   | 32 (72.7)                                  | 168 (81.2)                                     |         |
| Diabetes mellitus, n (%) |                                        |                                                 |         |
| Yes                  | 5 (11.4)                                   | 19 (9.2)                                       | 0.584   |
| No                   | 39 (88.6)                                  | 188 (90.8)                                     |         |
| Hypertension, n (%)  |                                            |                                                 |         |
| Yes                  | 18 (40.9)                                  | 58 (28)                                        | 0.091   |
| No                   | 26 (59.1)                                  | 149 (72)                                       |         |
| Dyslipidemia, n (%)  |                                            |                                                 |         |
| Yes                  | 39 (88.6)                                  | 152 (73.4)                                     | 0.032   |
| No                   | 5 (11.4)                                   | 55 (26.6)                                      |         |
| BMI, kg per m²       | 27.6 ± 4.0                                 | 26.2 ± 3.8                                     | 0.048   |
| Obesity, n (%)       |                                            |                                                 |         |
| Yes                  | 33 (75)                                    | 129 (62.3)                                     | 0.110   |
| No                   | 11 (25)                                    | 78 (37.7)                                      |         |
| Waist circumference, cm | 97.6 ± 10.7                                | 92.4 ± 9.7                                     | 0.004   |
| Central obesity, n (%) |                                        |                                                 |         |
| Yes                  | 36 (81.8)                                  | 151 (72.9)                                     | 0.220   |
| No                   | 8 (18.2)                                   | 56 (27.1)                                      |         |
| SBP, mmHg            | 139 ± 15                                   | 133 ± 14                                       | 0.024   |
| DBP, mmHg            | 82 ± 10                                    | 79 ± 11                                        | 0.085   |
| FBS, mmol/L          | 5.4 ± 0.9                                  | 5.4 ± 1.4                                      | 0.994   |
| TC, mmol/L           | 6.0 ± 1.1                                  | 5.4 ± 1.1                                      | 0.005   |
| HDL, mmol/L          | 1.4 ± 0.3                                  | 1.4 ± 0.3                                      | 0.913   |
| LDL, mmol/L          | 3.3 ± 1.4                                  | 3.0 ± 1.2                                      | 0.130   |
| TG, mmol/L           | 1.8 ± 0.8                                  | 1.7 ± 1.0                                      | 0.224   |
| ALT, IU/L            | 40 (32–43)                                 | 32 (23–39)                                     | <0.0001 |
| AST, IU/L            | 36 (24–40)                                 | 25 (20–34)                                     | 0.001   |
| GGT, IU/L            | 59 (35–73)                                 | 43 (26–59)                                     | 0.001   |
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There were more subjects with NAFLD with an increased CIMT compared to those without NAFLD. However, the difference was not statistically significant, likely due to the small number of subjects with ultrasound carotid Doppler in our study. In a large study on male subjects undergoing health screening in South Korea, persistent NAFLD based on ultrasonography was

Table 6  Characteristics of participants with and without raised CIMT

|                          | Participants with increased CIMT, n = 38 | Participants without increased CIMT, n = 93 | P-value |
|--------------------------|------------------------------------------|---------------------------------------------|---------|
| Age, years               | 55.1 ± 6.6                               | 46.3 ± 10.5                                 | <0.0001 |
| Gender, n (%)            |                                          |                                             |         |
| Male                     | 34 (89.5)                                | 65 (69.9)                                   | 0.018   |
| Female                   | 4 (10.5)                                 | 28 (30.1)                                   |         |
| Ethnic group, n (%)      |                                          |                                             |         |
| Malay                    | 19 (50.0)                                | 57 (61.3)                                   | 0.152   |
| Chinese                  | 6 (15.8)                                 | 14 (15.1)                                   |         |
| Indian                   | 13 (34.2)                                | 22 (23.6)                                   |         |
| Smoking, n (%)           |                                          |                                             |         |
| Yes                      | 6 (15.8)                                 | 15 (16.1)                                   | 0.962   |
| No                       | 32 (84.2)                                | 78 (83.9)                                   |         |
| Diabetes mellitus, n (%) |                                          |                                             |         |
| Yes                      | 10 (26.3)                                | 5 (5.4)                                     | 0.001   |
| No                       | 28 (73.7)                                | 88 (94.6)                                   |         |
| Hypertension, n (%)      |                                          |                                             |         |
| Yes                      | 20 (52.6)                                | 24 (25.8)                                   | 0.003   |
| No                       | 18 (47.4)                                | 69 (74.2)                                   |         |
| Dyslipidemia, n (%)      |                                          |                                             |         |
| Yes                      | 31 (81.6)                                | 68 (73.1)                                   | 0.367   |
| No                       | 7 (18.4)                                 | 26 (26.9)                                   |         |
| BMI, kg per m²           | 27.7 ± 3.8                               | 26.8 ± 3.7                                  | 0.203   |
| Obesity, n (%)           |                                          |                                             |         |
| Yes                      | 29 (76.3)                                | 68 (73.1)                                   | 0.705   |
| No                       | 9 (23.7)                                 | 25 (26.9)                                   |         |
| Waist circumference, cm  | 98.7 ± 7.2                               | 94.4 ± 9.2                                  | 0.006   |
| Central obesity, n (%)   |                                          |                                             |         |
| Yes                      | 36 (94.7)                                | 78 (83.9)                                   | 0.150   |
| No                       | 2 (5.3)                                  | 15 (16.1)                                   |         |
| SBP, mmHg                | 142 ± 16                                 | 133 ± 14                                    | 0.005   |
| DBP, mmHg                | 83 ± 10                                  | 79 ± 11                                     | 0.075   |
| FBS, mmol/L              | 6.0 ± 1.7                                | 5.3 ± 1.1                                   | 0.004   |
| TC, mmol/L               | 5.7 ± 1.4                                | 5.3 ± 0.9                                   | 0.098   |
| HDL, mmol/L              | 1.3 ± 0.3                                | 1.4 ± 0.3                                   | 0.757   |
| LDL, mmol/L              | 3.3 ± 1.4                                | 2.7 ± 1.2                                   | 0.029   |
| TG, mmol/L               | 1.7 ± 0.6                                | 1.7 ± 1.3                                   | 0.636   |
| ALT, IU/L                | 38 (25–47)                               | 30 (23–39)                                  | 0.026   |
| AST, IU/L                | 26 (21–36)                               | 25 (20–35)                                  | 0.711   |
| GGT, IU/L                | 49 (27–65)                               | 42 (23–59)                                  | 0.197   |
| NAFLD, n (%)             |                                          |                                             |         |
| Yes                      | 29 (76.3)                                | 55 (59.1)                                   | 0.063   |
| No                       | 9 (23.7)                                 | 38 (40.9)                                   |         |
| Advanced fibrosis, n (%) |                                          |                                             |         |
| Yes                      | 7 (18.4)                                 | 16 (17.2)                                   | 0.868   |
| No                       | 31 (81.6)                                | 77 (82.8)                                   |         |
| WHO recommendation on physical activity for health achieved (assessed using the GPAQ), n (%) | | | |
| Yes                      | 31 (81.6)                                | 76 (81.7)                                   | 0.985   |
| No                       | 7 (18.4)                                 | 17 (18.3)                                   |         |

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; CIMT, carotid intima-media thickness; DBP, diastolic blood pressure; FBS, fasting blood sugar; GGT, gamma-glutamyl transferase; GPAQ, Global Physical Activity Questionnaire; HDL, high-density lipoprotein; LDL, low-density lipoprotein; NAFLD, non-alcoholic fatty liver disease; SBP, systolic blood pressure; TC, total cholesterol; TG, triglyceride; WHO, World Health Organization.
reported to be associated with the development of subclinical carotid atherosclerosis.\textsuperscript{23} Similar reports demonstrating the association between NAFLD and CIMT have been published.\textsuperscript{6,7,24–28} However, negative studies on the association between NAFLD and CIMT have also been reported.\textsuperscript{29,30} These studies were conducted among patients with diabetes mellitus. In a previous study on patients with diabetes mellitus at our center, NAFLD diagnosed by ultrasonography was reported not to be associated with ischemic heart disease.\textsuperscript{31} We hypothesize that the association of NAFLD and cardiovascular disease is more attenuated when there are more established risk factors for cardiovascular disease that are closely related to NAFLD.

Interestingly, we found that advanced liver fibrosis was not associated with an increased CIMT. This lack of association was regardless of whether the 8 kPa or 9.6 kPa cut-off value was used for the diagnosis of advanced fibrosis (data not shown). One possible explanation is that the development of cardiovascular disease and the progression of liver disease in subjects with NAFLD occur through separate pathways, which may be differentially activated in each individual, thus accounting for the varied outcome. Cusi described the current understanding of the pathophysiology of NAFLD, metabolic syndrome, and cardiovascular disease.\textsuperscript{32} To the best of our knowledge, this is the first population-based study that evaluated the association between advanced liver fibrosis and subclinical atherosclerosis in the context of NAFLD. In the present study, the factor associated with increased CIMT was older age in concordance with previous community studies.\textsuperscript{33,34}

There were several strengths to this observational study. First, it was conducted among health-screening subjects and not among healthcare-seeking adults. Hence, the data are a close estimate of prevalence in a population-based setting. Second, as mentioned before, a more sophisticated and accurate method of detecting NAFLD and liver fibrosis with transient elastography was utilized for the first time in this study, compared to other population-based studies on NAFLD. The CAP and LSM are reliable in the evaluation of significant hepatic steatosis and advanced fibrosis, respectively, and, more importantly, are practical and acceptable to the subjects for the purpose of this study.\textsuperscript{3,4}

This study had several limitations. First, due to the sample size of the patients with raised CIMT, we could not draw conclusions on the association of NAFLD with increased CIMT. A larger number of study subjects would be needed to address this limitation. Moreover, the study participants were predominantly middle-class income people, and therefore, this study may not be representative of the general population. The reliability criteria for CAP were not yet established at the time this study was conducted and completed, and therefore, it was not applied in this study. In addition, NAFLD and raised CIMT share common risk factors, such as hypertension, dyslipidemia, and type 2 diabetes. These can be confounding factors in the analysis of the association between NAFLD and raised CIMT. Furthermore, the cross-sectional study design only enabled us to study an association between epidemiological factors with NAFLD and CIMT, rather than causation, which would have been better evaluated using a longitudinal study design.

In conclusion, the prevalence of NAFLD and advanced liver fibrosis was alarmingly high in this middle-aged multiethnic Malaysian population that attended a health screening. NAFLD, advanced liver fibrosis, and increased CIMT were found to be associated with traditional risk factors. NAFLD and advanced liver fibrosis appeared not to be associated with increased CIMT. However, a larger sample size is needed to demonstrate whether there is any association.

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